Completion Report

Eisenhower Consortium for Western Environmental

Forestry Research

Grant #EC401

Effects of Prescribed Fire on Water-Soluble Nutrients and Organics

and

Effects of Volatiles on Nitrogen Mineralization

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Acknowledgements

The Eisenhower Consortium for Western Environmental Forestry Research (Grant #EC-401) provided the majority of the funding with additional funding from the National Science Foundation (Grant #BSR-8312672). Personnel of the Santa Fe National Forest and particularly the Jemez Ranger District provided necessary information and assistance. They performed the prescribed burns and provided calculations on fire behavior. Without their assistance, this research could not have been performed.

Introduction

This report is the final project report for The Eisenhower Consortium for Western Environmental Forestry Research Grant #EC-401. The original grant was titled "Effects of prescribed fire on water-soluble nutrients and organics". Since the processes of nitrogen mineralization and nitrification determine the amounts of inorganic nitrogen produced from organic matter, prescribed fire would have to alter these processes if changes in water-soluble nutrients (particularly nitrate) would occur. As our research progressed, water-soluble and volatile organics appeared to play major roles in determining the rates of ammonification (the first inorganic nitrogen form in the process of nitrogen mineralization) and nitrification in this ecosystem. Amendments to the original proposal allowed us to pursue the effects of organics on nitrogen mineralization and nitrification. Thus, it seemed reasonable to arrange the final report into three sections: 1) Effects of Fire on Nitrogen Mineralization and Nitrification; 2) Effects of Fire on Organics; and 3) Effects of Volatiles on Nitrogen Mineralization. Each section is separate with the exception of the Literature Cited section which includes citations from all three sections.

Table of Contents

Section 1

Effects of Fire on Nitrogen Mineralization and Nitrification

Introduction

Nitrogen is often the element limiting productivity of terrestrial ecosystems. The majority of the nitrogen which is available for plant uptake is supplied by the complex process of nitrogen mineralization. This process converts organic nitrogen to inorganic nitorgen and can limit the rate of terrestrial nitrogen cycles. The process of ammonification produces the first inorganic form of nitrogen (ammonium) in the process of nitrogen mineralization. Measurements of nitrogen mineralization rates include the production of ammonium and nitrate unless steps are taken to prevent mineralized ammonium from being converted to nitrate by the two-step process of nitrification. All of these processes and the factors controlling them have received considerable attention (e.g., Clark and Rosswall 1981).

Ponderosa pine ecosystems have been identified as having low nitrogen mineralization rates and low nitrification rates in both laboratory and field studies (Powers 1980; Vitousek et al. 1982; Lodhi and Killingbeck 1980). Lodhi and Killingbeck (1980) and Vitousek et al. (1982) have identified many factors which could limit nitrogen mineralization and/or nitrification rates in ponderosa pine ecosystems. However, none of these factors have been directly tested in the field. The only tests were made using laboratory cultures of a single species of nitrifying bacteria.

Recent management of ponderosa pine ecosystems has increased the use of prescribed fire. The primary use of prescribed fire is to reduce the accumulated fuels in these ecosystems. Klemmedson (1976) expressed the concern that fire in these ecosystems may reduce the easily decomposed fraction of the forest floor and leave the fraction more resistant to

decomposition. This in turn may have detrimental effects on the ability of the ecosystem to provide inorganic nitrogen via the process of nitrogen mineralization.

To test the hypothesis that prescribed fire reduces decomposition and nitrogen mineralization, we collected samples of forest floor and mineral soil from areas recently managed with prescribed fire and adjacent unburned areas. The preliminary data from these samples indicated the rates of both mineralization and nitrification may be increased, not decreased, by prescribed fire. But perhaps of greater importance, these data suggested that soil obtained after prescribed fire was valuable in testing any hypothesis concerning inhibition of mineralization and/or nitrification processes. The inherently low rates of ammonification and nitrification in materials from natural ponderosa pine make it difficult, if not impossible, to test factors responsible for the inhibition of these processes. Prescribed fire may increase these process rates enough to allow detection of inhibition in a bioassay experiment.

This study was undertaken to determine the effects of prescribed fire on rates of nitrogen mineralization and nitrification of forest floor and mineral soil in a ponderosa pine ecosystem. In addition, we were hopeful that this study would provide some insight into the mechanism(s) that controls these processes. If prescribed fire resulted in increased rates of mineralization or nitrification, then we proposed to perform a bioassay experiment. The bioassay would use the mineral soil after prescribed fire to test the current hypotheses concerning allelochemic inhibition of these processes.

Literature Review

A ponderosa pine stand in New Mexico was one of 17 forested sites studied by Vitousek et al. (1982). Their research identified processes having the potential to control the loss of nitrate after disturbance of forest ecosystems. Field trenching experiments identified the ponderosa pine site as having: 1) a lag in the production of inorganic ammonium (the first inorganic form of nitrogen in the process of nitrogen mineralization), and 2) a lag in the conversion of inorganic ammonium to nitrate (the process of nitrification). These field results were also seen in laboratory measurements of nitrogen mineralization potentials (Fig. 1.1). Laboratory incubations of soil collected in different seasons revealed differing lag times in the production of both ammonium and nitrate. The three collections shown in Fig. 1.1. demonstrate the greatest range in lag time and production rates found in the ponderosa pine site.

Both the field trenchng experiment and the nitrogen mineralization potential measurements could underestimate the gross amount of mineralized nitrogen if gaseous loss was significant. The two processes which result in gaseous loss of nitrogen are ammonium volatilization and denitrification. Gosz and White (unpublished data) performed ^{15}N tracer experiments to determine the amount of gaseous loss from the incubation technique. They reported 100% recovery of the added ^{15}N from soils after incubation for 10 weeks. Thus, we assume the nitrogen mineralization potential measurement was not subject to gaseous loss for samples from the ponderosa pine ecosystem.

Many factors were identified by Vitousek et al. (1982) which could limit nitrogen mineralization and nitrification rates. These limiting factors include: 1) nitrogen availability, 2) macro- or micronutrient availability

other than nitrogen, 3) low field microbial populations, 4) high C/N ratio substrate resulting in immobilization of nitrogen in rapidly growing microbial populations, 5) organic substrate which is slowly mineralized due to its refractory nature (independent of C/N ratio), and 6) allelochemic inhibition of microbial processes by either direct toxic effects or by reducing access to otherwise readily decomposable organic substrate.

The majority of these factors were tested by Gosz and White (unpublished data) on samples from ponderosa pine. Their results are given in Table 1.1. All amendments containing inorganic ammonium resulted in significantly higher levels of nitrate while only the amendment with ammonium and sterilized soil inoculum significantly increased ammonium levels. However, when the amount of nitrogen added in the amendment was substracted from the final nitrogen levels, none of the amendments significantly increased the net amount of inorganic nitrogen mineralized from the organic substrate in the soil.

These results excluded all but three factors which could account for the low nitrogen mineralization rates and the lag in nitrification in the soil from this ponderosa pine site: 1) microbial immobilization of inorganic nitrogen (high C/N ratio organic substrate), 2) refractory organic substrate, and 3) allelochemic inhibition. Vitousek et al. (1982) could not predict nitrogen mineralization rates from the C/N ratio of the substrate in the soils of 17 sites ($r^2 = 0.03$). This suggests immobilization did not control nitrogen mineralization in the sites they studied. Thus, they concluded that sites with low mineralization rates (including the ponderosa pine site) had the following characteristics: 1) litter which is refractory to decomposition and also has a wide C/N ratio, and 2) the lag in nitrification could be caused by allelochemic inhibition of nitrifiers or by other mechanisms which could

not be separated by their data. Their analyses could not determine whether refractory organic substrate or allelochemic inhibition was the factor controlling mineralization rates in the ponderosa pine site.

Allelochemic inhibition has been suggested in other coniferous systems to control mineralization rates (Lamb 1975, Gosz 1981). It is often suggested that polyphenols of the litter are the allelochemic agents. Gosz (1981) reviewed the mechanisms by which polyphenols can reduce litter decomposition rates. These included: 1) reaction with plant proteins to produce resistant complexes, 2) coating or encrusting decomposable plant substances with resistant polyphenol complexes, 3) complexing of plant cellulose and hemicellulose with polyphenols or their absorption of polyphenols, and 4) reaction with microbial enzymes.

Allelochemic inhibition of nitrification and nitrifying bacteria in a ponderosa pine ecosystem was reported by Lodhi and Killingbeck (1980). They suggested the inhibition was caused by secondary plant chemicals (polyphenols in general, especially condensed tannins) produced by ponderosa pine. Water-extracts of ponderosa pine needles had the highest polyphenol content and the greatest toxic effects on nitrifying bacteria (a 93% reduction in the number of nitrifying bacteria in laboratory cultures). The reduction in number of bacteria was attributed to the direct toxic effects of the extracted compounds.

Klemmedson (1976) suggested fire in ponderosa pine ecosystems may reduce the easily decomposed fraction of the forest floor and leave the fraction more resistant to decomposition. This hypothesis is compatible with Vitousek *et al*. (1982) and assumes refractory and/or high C/N material is responsible for the low mineralization rates in ponderosa pine ecosystems. Although there are

review articles on the effects of fire on the nitrogen cycle (e.g., Raison 1979) and on fire effects in ponderosa pine (e.g., Wright 1978, Wright and Bailey 1982), we are unaware of a study which has directly measured decomposition of residual forest floor or soil organic matter after prescribed fire in ponderosa pine. Covington (1978) found a 20% reduction in forest floor organic matter (2.0 mm and smaller fraction) immediately after a prescribed fire in a ponderosa pine ecosystem in Arizona. His measurements eight months later found another similar reduction in the forest floor, presumably the result of increased microbial decomposition of the residual organic matter.

The total amount and availability of carbon are central to the current hypotheses on the mechanisms controlling nitrogen mineralization in ponderosa pine ecosystems. Thus, it would be beneficial to have a direct measure of the rate of carbon mineralization. Vitousek et al. (1982) did not measure carbon mineralization on material from ponderosa pine nor has anyone else to my knowledge. The rate of CO_2 evolution from soils has been found to accurately reflect the amount of organic matter mineralized during decomposition by heterotrophic organisms (Atlas and Bartha 1981). This measurement is often termed heterotrophic respiration. If a large quantity of easily decomposed organic matter with a high C/N ratio exists in the forest floor and/or mineral soil of ponderosa pine, then a relatively high rate of respiration would occur in these materials. If the organic matter in the ponderosa pine ecosystem is relatively recalcitrant in nature, the rate of respiration would be low when expressed on a per gram organic matter basis. In addition, if fire were to reduce the amount of easily decomposed material, the rate of respiration would be reduced when expressed on a per gram of organic matter basis.

Thus, measurements of respiration and nitrogen mineralization could perform the dual function of identifying the mechanism(s) controlling nitrogen mineralization rates and identifying the effects of fire on the rate of decomposition of the residual organic matter. The only mechanisms these measurements would not be able to separate are refractory organics and allelochemic inhibition. Readily decomposable material when in the presence of inhibitory allelochemics would result in the measurement of low respiration rates as would recalcitrant materials. The organic matter in ponderosa pine would have to be altered in some manner to achieve higher respiration and/or higher nitrogen mineralization and nitrification rates if inhibition of these processes were to be detected. Based upon the initial results, prescribed fire may provide just such organic matter.

Experimental Design

The problem considered in this section had three objectives: 1) determine if the organic matter not consumed by a prescribed fire treatments had a lower rate of decomposition than the original organic matter; 2) determine if the nitrogen mineralization rate of the forest floor and soil was altered by the prescribed fire treatment; and 3) if the nitrogen mineralization rates were increased by the fire treatment, perform a bioassay to determine if water-soluble components of the forest floor inhibited nitrogen mineralization and/or nitrification. The first two objectives were directly related and comprised the first phase of the study. The third objective was only performed after a significant increase in the mineralization and nitrification rates were found in the forest floor.

The first objective was designed to test the concern expressed by Klemmedson (1976) that prescribed fire in ponderosa pine may leave the organic matter which is more resistant to decomposition. The rate of CO_2 evolution is a measure of the rate of decomposition. If the rate was lowered, then Klemmedson's hypothesis would be supported. It was essential to express these data on an orgainc matter basis (ash-free weight) to test the hypothesis since the fire treatment would lower the amount of organic matter and increase the amount of mineral material.

The second objective of the study was directly related to the first. The net result of decomposition is the release of inorganic nutrients. A decreased rate of decomposition would decrease the rate of inorganic nitrogen production. Since the fire process directly releases inorganic nutrients, it was necessary to separate this production from later production. Thus, it was essential to collect the forest floor and mineral soil immediately before and

after the fire to measure the amount of inorganic nitrogen mineralized by the fire treatment. This initial mineralization would then be subtracted from subsequent inorganic nitrogen levels to determine the net rate of nitrogen mineralization. If nitrogen mineralization and nitrification would be lowered by a prescribed fire treatment, Klemmedson's hypothesis would be supported. If nitrogen mineralization and nitrification would be increased, the third objective of the study would be performed.

The third objective of the study was dependent upon finding increased rates of nitrogen mineralization and/or nitification after the prescribed fire treatment. If the rates were increased, some property of the forest floor or soil had to have been altered by the fire treatment. Either 1) an inhibitor was removed, combusted, or inactivated, 2) immobilization of nitrogen was reduced by a concurrent reduction in the amount of readily avaliable carbon substrate, 3) previously unavailable or slowly available substrate was made more readily available, or 4) a combination of two or more of these mechanisms. Regardless of the alteration, material supporting increased mineralization and nitrification was essential for testing the hypotheses concerning inhibition of mineralization and/or nitrification in natural ponderosa pine ecosystems.

The literature suggests the three mechanisms which most likely control nitrogen mineralization and nitrification rates in ponderosa pine ecosystems are: 1) high C/N ratio substrate which is available to microbial decomposers resulting in immobilization of nitrogen in rapidly growing microbial populations; 2) organic substrate which is slowly mineralized due to its refractory nature (independent of C/N ratio); and 3) allelochemic inhibition of microbial processes either by direct toxic effects or by reducing access to

otherwise readily decomposable organic substrate. For the bioassay to separate these mechanisms, it was essential to measure both heterotrophic respiration and nitrogen mineralization on the same assay material. Figure 1.2 may aid in understanding this need. The application of the water-extracts could cause a delay in ammonification and nitrification as a result of two very different mechanisms which are related to the availability of the carbon substrate: 1) application of an inhibitor (treatment in Fig. 1.2) would result in lowered activity by the decomposer organisms, thus lowering the processing rates and lengthening the time scale for the production of inorganic ammonium; or 2) application of readily available organic substrate in the water-extracts (treatment in Fig 1.2) would result in increased growth in the microbial populations, increased respiration, and microbial immobilization which would delay the production of inorganic ammonium until microbial growth requirements for nitrogen were satisfied. An increase in the respiration rate would indicate the application of readily available substrate (such as sugars). By measuring heterotrophic respiration and nitrogen mineralization potential, information about the mechanism(s) controlling nitrogen mineralization in ponderosa pine ecosystems could be obtained.

The null hypothesis for the bioassay was:

H_o: The rate of heterotrophic respiration, nitrogen mineralization, and nitrification in the assay soil was not altered by the application of water extracts from the forest floor.

No change in any of the measured rates would support the null hypothesis. This would support the hypothesis that refractory organic substrate which is resistant to decomposition controls nitrogen mineralization. A number of

other results were possible. A list of the possible results along with our interpretation included:

1) The control (unburned) forest floor extract lowered CO_2 evolution and lowered nitrogen mineralization while the burned forest floor extract did not significantly alter these processes. Allelochemic inhibitors of decomposition and nitrogen mineralization were extracted from the control forest floor. The prescribed fire treatment combusted or inactivated these inhibitors.

2) The control forest floor extract increased CO_2 evolution and lowered nitrogen mineralization rates. High C/N ratio material caused microbial immobilization of nitrogen. The fire treatment probably combusted or volatilized the high C/N ratio compounds which resulted in the higher nitrogen mineralization rate in the assay soil.

3) The control forest floor extract and the burned forest floor extract had the same rates of nitrogen mineralization, but the control forest floor extract produced proportionally more ammonium and less nitrate relative to the burned forest floor extract. Nitrification inhibitors were extracted from the control forest floor (supports Lodhi and Killingbeck 1980). Prescribed fire either combusted or altered the inhibitors.

Combinations of these results were possible. For example, the addition of an inhibitor along with the addition of readily available substrate in the water-extracts may cause off-setting measurements. We anticipated the results may not be clear-cut, but valuable information and insight may be obtained.

Site Description

A site in the Jemez Mountains near Bear Springs, New Mexico, was selected for this study. The elevation is 2225 m. The ponderosa pine stand is on fairly uniform volcanic tuff. The 0-10 cm depth soil (<2.0 mm diameter) is a sandy loam with an average composition of 56.6% sand, 27.6% silt, and 15.8% clay. The amount of sample between 2 and 6.4 mm diameter (a 6.4 mm sieve was used to maintain soil structure in all field and laboratory incubations) averaged 13.3% of the total sample. Bulk density averaged 0.899 g cm^{-2}. The area had been logged in the 1930's. None of the larger trees left at that time were within the study plots and their buffer zones. The average diameter at breast-height (DBH) for all trees within the plots and buffer areas was 9.4" and the average age measured at breast-height was 51 years. The regression of age and DBH was not significant (r^2=0.33). The area was designated to be managed with prescribed fire. The fuels ranged from about 2 to 17 tons per acre with the forest floor (litter and duff; or the L, F, and H horizons) ranging from <4 to about 16 tons per acre.

Methods

It was essential to obtain pre-treatment measurements on the proposed treatment and control plots to establish the relationship between the proposed treatment and control plots. The effects of the prescribed fire treatments could only be determined knowing the pre-treatment relationships.

Eight plots measuring 4 m X 9 m with a 5 m buffer zone around the perimeter were established. The four plots to receive the fire treatment were chosen at random with the other four used as controls. All plots had fairly uniform forest floors averaging 11 tons $acre^{-1}$ of fine fuels (<1/4") and 2.5 tons $acre^{-1}$ of larger fuels. Heavy fuel deposits were avoided. Each 4 m X 9 m plot contained ten line transects 4 m long placed at 1 m intervals. A single line transect was chosen at random for each collection and the same numbered transect was collected at each plot.

Forest floor samples contained all organic horizons (L, F, and H). The entire forest floor beneath a 100 cm^2 template was collected (Gosz et al. 1976). Five replicates were taken at 1 m intervals along the selected line transect. The five replicates were composited.

Mineral soil samples were collected with a 10 cm diameter auger to a 10 cm depth. The collection was made at the same point along the line transect where the forest floor was collected. The five replicates were composited in the field. Roots were removed by hand sorting. Stones larger then 1/4" were removed by sieving. Subsamples of 10 g of this composited mineral soil were placed in five preweighed bottles, each containing 100 ml of 2 N KCl (with PMA preservative), and five preweighed soil cans. Five 100 g subsamples (at field moisture content) from each plot were placed in plastic sandwich bags and sealed with twist-ties. One was returned to each of the five points of soil

collection and covered with nearby soil. These buried bags were used to measure field mineralization. The remainder of the composited mineral soil was transported to the laboratory.

In the laboratory, twigs and stones larger than 1/4" diameter were removed from forest floor collections, dried to constant weight at 105^{o}C, and the weight recorded. The remainder of the forest floor and mineral soil composites were subsampled for measurement of moisture content, total N and P, and ash content. The water content at -0.1 bars was determined for each composite by applying suction to a soil pressure plate holding a water saturated subsample of the composite. Each composite was then adjusted to -0.1 bars and this material was then used for laboratory determination of nitrogen mineralization potential and potential CO_2 respiration rates.

The method of Vitousek et al. (1982) was used to determine the potential rate of nitrogen mineralization. This method used aerobic incubation of samples at 20^{o}C. A total of 35 subsamples of each composite were weighed into plastic cups. Five cups of each composite were immediately extracted with 100 ml of 2 N KCl (with PMA preservative). Five cups of each composite were extracted after incubation of 1, 2, 4, 6, 8, and 10 weeks. The clarified extracts were analyzed for inorganic ammonium by the modified Technicon AutoAnalyzer method of White and Gosz (1981) and for nitrate by the Technicon AutoAnalyzer cadmium reduction method.

Potential respiration rates were measured by the alkaline trap method. Fifty g of mineral soil or 15 g of forest floor were placed in 500 ml glass jars and the total weight recorded on the jar. A total of three replicate jars were used for each composite. An open scintillation vial containing 5 ml

of 1 N NaOH was placed in each jar and the sealed jar incubated at 20°C. The vials containing the NaOH were removed at the same intervals as the cups and replaced with vials containing fresh NaOH. The jars were adjusted to their initial water content when needed. The removed NaOH was quantitatively transferred to a 150 ml beaker, 5 ml of 1 N $BaCl_2$ added to precipitate the diffused CO_2 as insoluable $BaCO_3$, and titrated to a pH of 8.3 with 0.1 N acid phthalate. A jar containing only the NaOH trap was treated in the same manner as the samples and provided an experimental blank to correct for absorbed atmospheric CO_2. The amount of CO_2 liberated from the sample was calculated as follows:

$$\text{mg } CO_2 = 22 \text{ (ml to titrate blank - ml to titrate sample)}$$
$$\text{X (molarity of acid)}$$

Field nitrogen mineralization was determined with a buried bag technique. The bottles and soil cans containing mineral soil samples from the initial field collection were reweighed in the lab to determine the field weight of the each sample. The soil cans were dried to constant weight at 105°C. An average moisture correction factor was applied to the field weight of the KCl extracted samples to obtain oven dry weight. A clarified portion of the KCl extract was analyzed for inorganic ammonium and nitrate as described above. These values provided initial ammonium, nitrate, and moisture content for the samples in the buried bags. At the next collection, the bags were retrieved. After mixing, a portion of each sample was placed in soil cans and preweighed bottles containing 100 ml of 2N KCl. These cans and bottles were treated as described above. This procedure gave the initial inorganic nitrogen values and the values after the period of incubation in the field.

Moisture content was determined by weight loss upon heating at 105^{o}C to constant weight. Total N and P were determined by a modified Kjeldahl digestion (Schuman et al. 1973) followed by analysis for ammonium and orthophosphate on a Technicon AutoAnalyzer. Ash content was determined by loss upon ignition at 500^{o}C.

Bioassay Methods

The immediate post-burn samples identified plot #7 as having significantly increased mineralization and nitrification rates in the mineral soil. Five months after the fire treatment, five samples of forest floor and mineral soil were taken from the perimeter of the 4 X 9 m area of plot #7 (treated with prescribed fire) and forest floor from plot #8 (a control plot) using the methods described above. The composite mineral soil from plot #7 was used as the assay soil.

The forest floor composites from plot #7 and #8 were extracted with 1250 ml of water. This amount simulated a 1" precipitation event completely passing through the composited forest floor samples. After agitation by hand, the samples were allowed to settle overnight and then the forest floor solution was poured off and suction-filtered. Both extracts were diluted by 5 and by 25 to give 1X, X/5, and X/25 concentrations.

The bioassay experiment consisted of the application of each of the extract solutions (including the dilutions) and a demineralized water blank solution to the assay soil. The appropriate amount of an extract was added to a subsample of the assay soil to simulate the 1" precipitation event added to the top 10 cm of mineral soil. After addition of the extract solution, the mineral soil subsample was adjusted to a water content of -0.1 bars.

Measurements of nitrogen mineralization potential and respiration were performed on each subsample using the above methods.

Statistical Methods

All statistical analyses were performed on SAS programs (Statistical Analysis System, SAS Institute Inc. 1982). ANOVA, MANOVA, and Duncan's Multiple Range Test were used for most analyses. The 95% confidence level was used for all tests of significance ($P < 0.05$). The mean of the replicate analyses for each measurement for each plot was used in the statistical analyses to evaluate the effect of the fire treatment. This gave a sample size of 4 for each analysis for the weekly analyses. The total sample size to evaluate the effect of treatment was 28 and 24 for the nitrogen mineralization potentials and respiration analyses, respectively. The ANOVA or MANOVA analyses allowed the effect of treatment to be analyzed as a single factor. The weekly data were also analyzed for significant differences. This type of analysis allowed detection of significant treatment effects when none of the individual weekly analyses were significantly different.

Results

As stated earlier, it was essential to establish the degree of similarity between the proposed treatment and control plots prior to the application of the prescribed fire treatment. There were a total of 48 paired analyses for heterotrophic respiration (Table 1.2) and nitrogen mineralization potentials (Tables 1.3 and 1.4) on the samples from the three pre-treatment collections. Only seven of these analyses detected significant differences between the proposed burn and control plots. None of the analyses were significantly different in more than one of the three collections. Most importantly, none of the immediate pre-treatment analyses were significantly different.

Immediate Post-treatment Effects

The prescribed fire treatment was applied on November 7, 1983. The four plots and their surrounding buffer zones were burned in about 3 m wide strips. Specific climatic conditions and fire behavior characteristics are given in Table 1.5. The fire-line intensities were calculated to range from 38 to 59 BTU/ft second (or 131 to 203 kW/m). These were relatively cool fires and only burned to the mineral soil in isolated spots. Some spots (especially on plot #7 which was the last plot treated) remained too hot to sample on the day of the burn, thus, the post-treatment samples were collected the following morning (less than 24 hours after treatment). The burned soils (particularly plot #7) remained noticeably warmer than the unburned soils the following morning. There were no unusual climatic events nor was there any precipitation prior to collection of the post-treatment samples. The forest floor was reduced by an average of 44.6% (on an ash-free weight basis) while the total N was reduced by an average of 23.4%. The soils were not altered in their organic or total N content.

Field Nitrogen Levels

The amount of nitrogen mineralized in the forest floor by the fire treatments is given in Table 1.6. There was no significant difference in the treatment and control plots prior to the prescribed fire treatment. This pattern was true for all measurements and the discussion will concentrate on the difference between the pre- and post-burn collections from the treatment plots. The treatments significantly increased the amount of ammonium in the forest floor and siol while no significant change in nitrate was detected in either the forest floor or soil. The increase in ammomium was directly proportional to the forest floor consumption with correlation coefficients of 0.985 and 0.735 for the forest floor and soil, respectively. Soil from plot #7 (having the hottest treatment) displayed the largest increase in ammonium (1850%) immediately after the fire treatment.

Heterotrophic Respiration and Nitrogen Mineralization Potentials

The treatments significantly decreased respiration rates in the forest floor samples when expressed on a dry weight basis (Fig. 1.3). When these data are expressed on an ash-free weight basis (or as respiration per gram of residual organic matter), the weekly means were not significantly different (Fig. 1.4). There was a consistent trend of lower respiration rates in the burned forest floor samples. The ANOVA detected a significant treatment effect with the post-burn forest floor respiration rates significantly lower than the pre-burn but not different from the control forest floor (when expressed on an ash-free basis, Table 1.2). Soil respiration rates showed no significant treatment effect.

The nitrogen mineralizaton potentials of the pre- and post-burn forest floor samples are shown in Fig. 1.5. The mineralization potential of the pre-burn samples are rather typical for ponderosa pine forest floor (Vitousek

et al. 1982, Gosz and White in preparation). Ammonium levels decreased initially (immobilization phase) and then increased (mineralization phase) during incubation. Only a very slight increase in nitrate occurred during incubation. The post-burn samples displayed a much different response. The significant increase in ammonium which occurred immediately after the fire treatment can be seen in the post-burn week 0 level. Although ammonium displayed a similar trend as the pre-burn samples (first immobilization, then mineralization in later weeks), nitrate was significantly increased with production occurring within the first week of incubation. The increase in nitrate was nearly exponential throughout the incubation period, indicating substrate depletion did not occur during the 10-week incubation.

The nitrogen mineralization potentials of the pre- and post-burn mineral soil samples are shown in Fig. 1.6. The only significant effect the fire treatments had on these measurements was to significantly increase the initial ammonium levels (week 0). All other values were not significantly different during the entire incubation. These results are the means of all four treatment plots. There was a slightly different effect on each individual plot. The soils from plots #2 and #3 showed slightly decreased rates of nitrogen mineralization (although the decrease was not statistically significant). The soil from plot #6 was nearly identical before and after treatment. The soil from plot #7 showed a statistically significant increase in ammonium, nitrate, and net nitrogen mineralization immediately after the treatment. This plot had the highest calculated fireline intensity and perhaps the soil was heated more than the other soils. The soil from plot #7 was the ideal assay soil with increased rates of inorganic nitrogen production.

The immediate effects of the fire treatments are summarized in Table 1.7 (with the exception of the increase in soil ammonium levels only in week 0). The treatments significantly increased the initial inorganic nitrogen levels (all the increase occurred in the ammonium form), significantly increased the levels of inorganic nitrogen after 10-week incubation, and significantly increased the net mineralization rate in the forest floor. When these potential increases are expressed on an areal basis in the field, the net amount of inorganic nitrogen potentially available for plant uptake was almost tripled by the fire treatment.

Treatment Effects: Six-month Post-treatment Collection

The second post-burn collection was made on May 15, 1984. The winter and spring moisture during this six month interval was greater than average. In the May collection, heterotrophic respiration rates (expressed on an ash-free weight basis) of the forest floor samples from the burned plots were significantly lower than those from the control plots (Fig. 1.7). The control plots in the May collection had higher respiration than in the November collection (Table 1.2). The burned plots had slightly higher rates in May than immediately after the fire treatment in November (Table 1.2). The heterotrophic respiration rate in the mineral soil was nearly identical in the burned and control plots (Fig. 1.8) in the May samples.

The prescribed fire treatments significantly increased nitrogen mineralization in the forest floor (May 15,1984, Table 1.3). The only non-significant effect was on the ammonium levels expressed on a dry weight basis (although ammonium was higher in the burned plots). Forest floor nitrogen mineralization potentials in the May collection (Fig. 1.9) were similar to the November collection (Fig. 1.5). Initial ammonium levels were higher in samples from the burned plots than the control plots. Nitrate

production in the May collection was still significantly increased in the burned samples and was even greater than in the November collection. The control samples still had little nitrate production.

Although mean nitrogen mineralization in the soil was not significantly different immediately after the prescribed fire treatment, both nitrate production and net nitrogen mineralization (sum of ammonium and nitrate) were significantly increased in the May collection (on both dry weight and ash-free weight basis, see Table 1.4). Reasons for this delayed effect are discussed later. The nitrogen mineralization potentials for the soil samples in the May collection are shown in Fig. 1.10. The control plots were very similar to the pre-burn plots in the November collection (Fig. 1.6). In contrast, the burned plots started production of nitrate earlier and reached higher nitrate levels than the control plots.

Treatment Effects: Ten-months Post-treatment Collection

The third post-treatment collection was made on August 31, 1984 (about ten months after the treatments). Heterotrophic respiration rates were significantly lower in the burned plots than in the controls. However, the burned plots had almost the same rate of respiration as in the November and May samples when expressed on an ash-free basis. This suggests the decomposition rate of the burned forest floor did not change significantly after the initial response and the control forest floor was consistently increased in its potential decomposition rate. The soil respiration rates were not significantly different and were within the range of the other collections.

Nitrification rates in the forest floor were still significantly greater in the burned samples than in the control samples. Net mineralization rates were not significantly different although the burned forest floor samples were

nearly twice the control. Although the soils were not statistically different for any of the inorganic nitrogen levels, the burned soils were nearly twice the control soils for all levels. The reason these differences were not significant is probably due to the increased variation between the burned plots with plots #2 and #7 having even higher rates of mineralization and nitrification than in the earlier samples and plots #3 and #6 slightly greater or equal to the control plots.

Bioassay

All assay soils with forest floor extracts had significantly lower respiration rates than those of soil with the addition of demineralized water (water blank; shown as Water Blk in Table 1.8). During a single 2-week period of the incubation, the pH of the water blank soil was about 0.5 units lower than all the soils which received extract additions. The lower pH would result in a shift in the carbonic acid-bicarbonate equilibrium and result in the release of dissolved CO_2. Thus, some of the CO_2 could have been from this shift in dissolved CO_2 which is exogenous to respiration sources. The pH of all the other soils were within 0.2 units of each other throughout the incubation period. The addition of the most concentrated extracts (burned-1 and control-1) resulted in nearly equal heterotrophic respiration rates while the addition of all the dilutions resulted in lower heterotrophic respiration rates not significantly different from each other. This suggests that the most concentrated extracts increased the amount of available carbon relative to the dilutions. However, since the two concentrated extracts were not significantly different, they probably contained similar amounts of available carbon.

The forest floor extracts significantly altered the nitrogen mineralization potential of the assay soil (Table 1.9). All extracts resulted

in an increase in the mean ammonium levels in the soil relative to the demineralized water addition. Only the addition of the 1X and X/5 extracts resulted in nitrate amounts statistically equal to the water blank addition while all other extract additions resulted in decreased nitrate production. All extract additions, except the most dilute, resulted in net nitrogen mineralization rates (sum of ammonium and nitrate) significantly different from the water blank. However, net nitrogen mineralization was increased with the addition of extracts from the burned forest floor and decreased with the addition of extracts from the unburned forest floor.

The extracts were analyzed for major inorganic nutrients, pH, tanning capacity, and relative absorbance (Table 1.10). The concentrations of many nutrients varied between the two extracts. However, only 1.5 ml of the concentrated extract was applied to 10 grams (dry weight) of mineral soil and no change in soil nutrient levels or pH could be detected after application of the extracts. The tanning capacities of both extracts were at or below the detection limit. The relative absorbance of the extract from the burned forest floor was 70% of the extract from the control forest floor although the total weight of the burned forest floor was only 36.8% of the control. This suggests the majority of the water-soluble fraction of the forest floor was contained in the lower forest floor horizons which were the least consumed by the fire treatment. The humus layer of the forest floor typically contains large quantities of water-soluble "humic" compounds.

Field Mineralization

There were a number of problems with the field mineralization experiments. 1) The samples were buried at their current field moisture contents which was never constant between plots nor across collection periods. 2) Small invertebrates often penetrated the bags (either entering or exiting).

This resulted in these replicate soil samples having a different water content than the other replicates (sometimes drier, sometimes more moist). The openings also provided an entrance for roots which grew into the bags and may have depleted the inorganic nitrogen levels. 3) For some undetermined reason, some bags would accumulate moisture within the bag and the once nearly dry soil was nearly saturated with water. When this occurred and how long the soil was in this condition was impossible to determine. Each of these factors may have had a major impact on the amount of nitrogen mineralized during the field incubation and the amount remaining in the soil sample.

The mean quantities of nitrogen mineralized during the field incubations are given in Table 1.11. The incubation periods ranged from 77 days to 409 days. Some samples were incubated only during the warmer season, some were incubated only during the colder seasons, and others were incubated over an entire year. The effect of incubation temperature is obvious. The samples incubated in the field during the cooler months show lower mineralization rates or show net immobilization (negative mineralization values). The samples incubated during the warmer months were all higher when expressed on an annual basis. All the samples started the incubation at different moisture levels, but soil moisture was always lower than in the laboratory incubations. Although the field conditions (both moisture and temperature) were not optimal for nitrogen mineralization in the field, the results are very close to estimates from laboratory potential measurements. These data suggest the laboratory measurements over a 10-week incubation may closely approximate the amount of nitrogen mineralized in the field on an annual basis.

The field incubations did not show any statistically significant effect of the fire treatments. The variation between replicate bags within each plot was fairly large and the samples from different plots had different moisture

content at the start of the incubation and at the end. Thus, it is not really surprising the field incubations did not show any statistically significant effect. The field incubations did show some interesting trends. The proportion of nitrate relative to ammonium in soil samples from the burned plots was consistently greater than in samples from the control plots in the last field incubation. This may indicate a decrease in nitrification inhibitors in the burned plots relative to the control plots.

Discussion

The prescribed fire treatments were relatively light burns with low fireline intensities (Table 1.5). An average of 44.6% of the forest floor was consumed which was less than desired under the current burn prescription. Three of the burn treatments did not result in a significant reduction in soil moisture after the burn treatments (plot #7 was the only exception, see Table 1.5). This suggests the treatments did not generate enough heat to produce a significant heat pulse in the soil and to evaporate soil moisture. Plot #7 was the hottest of the burn treatments. This was caused by a number of contributing factors including: 1) lower forest floor moisture at the time of the burn, 2) higher air temperature, 3) lower relative humidity, 4) higher windspeed, and 5) the west facing aspect of the site. Field observations detected a considerable amount of smoldering, or glowing combustion, within the forest floor in plot #7 while the other plots had mainly flaming combustion with little smoldering. Glowing combustion is the dominant combustion process after the volatiles have been expelled from a cellulosic fuel leaving a shell of charcoal (Chandler et al. 1983). It appeared that relatively little charring of the residual forest floor occurred on all plots except #7.

Fire intensity is known to play a major role in determining the effects of any fire treatment (Chandler et al. 1983). The effect of the fire treatments were generally correlated with fireline intensity. When a given parameter was altered by the treatments, the plot showing the greatest change was plot #7 which received the highest intensity treatment. To what degree the results of this study are applicable to different intensity fires is unknown. We feel this study accurately reflect the effects of low-intensity

fire treatments, which perhaps were the types of fires the region experienced prior to fire exclusion.

The prescribed fire treatments resulted in a statistically significant decrease in heterotrophic respiration rates in the forest floor (Table 1.2). While the reduction was statistically significant, the actual reduction in respiration was only 20% and 23% in the November (immediate post-burn) and the May collections, respectively. Factors other than reduced decomposition rates could contribute to the reduction in CO_2 evolution: 1) increased demand for CO_2, and 2) a change in microbial respiration efficiency caused by a change in the composition of the heterotrophic soil community. The reduction in CO_2 evolution was accompanied by a highly significant increase in nitrate production. Nitrifying bacteria utilize dissolved CO_2 as their carbon source, but nearly 100 moles of nitrate production is required to fix one mole of CO_2 (Atlas and Bartha 1981). Consumption of carbon during nitrification could account for only a very small portion of the reduced CO_2 evolution. Different microbial communities have different respiration rates per unit of utilized substrate (Atlas and Bartha 1981). Since the mineralization and nitrification rates of the forest floor were changed by the fire treatment, it is reasonable to assume the composition of the microbial community also was changed. The reduction in respiration may represent a different respiration efficiency by a different microbial community in the forest floor after the fire treatments. This factor could explain the observed differences in respiration and merits further investigation.

Based on these results, we have to state that: Decomposition of the residual organic matter in the forest floor and soil was significantly lowered by the prescribed fire treatment. However, this statement is made rather

cautiously since respiration was only slightly lower and alternative explanations for the reduction are possible.

Nitrogen mineralization and nitrification were significantly increased in the forest floor immediately after the fire treatments and remained significantly higher 6 months after the fire. Initially, soil nitrogen mineralization was not changed by the fire treatments (except on plot #7), but both nitrification and net nitrogen mineralization were significantly increased 6 months after treatment (Table 1.4). This occurred although soil respiration was not significantly affected (although slightly increased, Table 1.2). Both nitrogen mineralization and nitrification potential of the forest floor and mineral soil were increased by the fire treatment.

Klemmedson (1976) thought prescribed fire in ponderosa pine would increase the proportion of organic matter resistant to decomposition. An increase in refractory materials would cause a reduction in the release of nitrogen from such materials when expressed on a total organic matter basis. This did not occur. Instead, lowered respiration rates in the forest floor accompanied higher nitrogen mineralization rates and nitrification rates. The following may explain these observations.

For a given amount of carbon mineralized during decomposition, a greater amount of inorganic nitrogen will be released from organic matter with a lower C/N ratio than from organic matter with a higher C/N ratio. Ponderosa pine litter (needles and woody material) has a very high C/N ratio (Vitousek et al. 1982). As this material decomposes, the C/N ratio becomes lower. This results in a C/N ratio gradient within the forest floor with the highest C/N ratio in the surface horizons and the lowest C/N ratio in the lower horizons (humus layers). The removal of the upper horizons should result in a lower C/N ratio in the residual forest floor. This was precisely the effect of the

fire treatments (carbon was lowered by 44.6% while nitrogen was lowered by only 23.4%). It appears the residual forest floor followed a decomposition pattern typical of lower C/N ratio organic matter and did not reflect a significant increase in the proportion of refractory materials. Thus, although the treatment caused a significant reduction in forest floor respiration, the reduction is unlikely to represent a major reduction in the decomposition rate of the residual organic matter.

The main objective of the practice of management with prescribed fire is to reduce the accumulated fuels and thus reduce wildfire hazards. The objective of reduced fuels and an increase in site fertility (measured by nitrogen mineralization rates) appears to be achieved fires which only partially consume the forest floor. Even fires with low consumption may increase the ability of the ecosystem to supply inorganic nitgrogen (and maybe other nutrients) in the immediate future.

Mineral soil from plot #7 was used for the assay soil in the bioassay experiment. It was noted earlier that the soil from this plot was the only soil to show an immediate increase in nitrification and mineralization rates. Also, this soil was the only soil to have a significant heat pulse (as shown by the reduction in soil moisture) and/or evaporation from the mineral soil after the fire. The heat pulse may have played an important role in the observed increase and will be discussed later. It is also important to note that the analyses of the mineral soil from plot #7 immediately before and after the prescribed fire treatment detected no change in soil pH or C/N ratio.

The water-extracts of the forest floor materials (burned and control) reflect the following commonly accepted effects of fire (Table 1.12): 1) an increase in pH, 2) an increase in base cations, and 3) an increase in nutrient

availability (particularly phosphorus). It is important to note that the addition of 1.5 ml of the water-extract did not alter the measured sample soil characteristics. Thus, the effects of the water-extracts can not be attributed to changes in major soil parameters (such as pH).

The bioassay did not detect any significant difference in the respiration rates of the concentrated extracts. The concentrated extracts did increase respiration relative to the diluted extracts; thus, the extracts apparently added an available carbon source. Since the assay soil was the same for both extracts, and since the extracts were filtered to exclude microorganisms, it is reasonable that the initial composition of the soil microbial community was the same immediately after the addition of the concentrated extract. If immobilization of nitrogen during utilization of the added carbon source controls nitrogen mineralization, then both extracts should have the same immobilization capacity since they exhibited the same amount of respiration. However, the concentrated extracts had opposite effects on nitrogen mineralization and nitrification. The control forest floor extract significantly reduced net mineralization and nitrification while the burned forest floor extract significantly increased ammonium production and net mineralization.

These results mean the null hypothesis must be rejected. An alternate hypothesis and explanation are needed. However, none of the originally anticipated results perfectly fit the observed patterns. The results suggest immobilization of nitrogen during decomposition of available high C/N ratio material does not control nitrogen mineralization processes in this soil since the extracts had the same measured immobilization capacity (alternate result #2 for bioassay). Also, the C/N ratio of the assay soil was not different than the soil before the fire treatment. Thus, the soil C/N ratio and

immobilization can probably be ruled out as mechanisms which control nitrogen mineralization in ponderosa pine soil.

Allelochemic inhibition of nitrification and net mineralization is strongly supported by these results. We propose that the fire treatment and the associated heat pulse in the soil inactivated inhibitors (denatured, heat decomposed, volatilized). This would account for the immediate increase in nitrogen mineralization and nitrification in both the forest floor and mineral soil in plot #7 after the fire treatment. The extract from the burned plot would contain little inhibitory material (unless critical amounts of the inhibitor were added in litterfall or throughfall during the winter). The addition of the burned plot extract should have primarily a fertilizer effect and stimulate nitrogen mineralization in the absence of an inhibitor. The control forest floor would not be depleted in inhibitory substances. The extract would contain any water-soluble inhibitor along with other organics. This explanation is consistent with the results.

The current theories concerning control of nitrogen mineralization and nitrification have centered on the effects of tannins and polyphenols (Lamb 1975, Gosz 1981, Rice 1974 and 1984, Lodhi and Killingbeck 1980). Inhibition of both ammonification and nitrification has been attributed to polyphenols and/or tannins. Lodhi and Killingbeck (1980) attributed the reduction in the number of laboratory cultured nitrifying bacteria to the direct toxic effects of water-extracted compounds in ponderosa pine needles. They concluded that those results support the hypothesis of inhibition of nitrification in climax ecosystems (Rice 1984) and suggested tannins as the likely inhibitory agent. The control forest floor extract definitely inhibited nitrification in the assay soil. Also, nitrification was inhibited in the nitrogen mineralization potentials of the original forest floor material from plot #8. However,

neither the extract nor the original forest floor material had significant tanning capacity (Table 1.12). Also, it is unlikely that the majority of the polyphenols and the compounds which contribute to the tanning capacity would have been significantly altered in the soil by the heat pulse. Inhibition of nitrogen mineralization processes can not be attributed to tannins in these samples.

Soil temperatures were not measured directly during or after the fire treatment. Maximum temperature and duration of heating of the soil is unknown; however, compounds with significant volatilization or boiling points around 100°C or lower should have been the most affected by the fire treatment. This fits the characteristics of the terpenoids rather than polyphenols. The role of volatile organics on nitrogen mineralization processes is unknown and will be the subject of the following sections.

It is important to note that the control forest floor extract also significantly reduced net mineralization, although to a lesser degree than nitrification. This is not consistent with the theory of inhibition of nitrification in climax ecosystems (Rice 1984). Central to this theory is that nitrogen cycling and nitrogen mineralization rates are near maximal in the climax ecosystems which exhibit inhibition of nitrification. The strategy is to retain the mineralized nitrogen in the ammonium form and prevent the transformation to nitrate (which can more easily be lost from the ecosystem). The inhibition of both nitrogen mineralization and nitrification by the addition of water-extracted forest floor material indicates ammonification may also be inhibited. The inhibitor(s) may be general microbial inhibitors and are merely more efficient at inhibiting heterotrophic nitrifiers.

The role of water-soluble (completely or partially soluble) inhibitors in controlling the processes of nitrogen mineralization and nitrification is

strongly supported by these results. The extracts simulated a single leaching of the forest floor by a 2.54 mm precipitation event. Beneath unburned forest floor, inhibitors may be contributed during each precipitation event which reaches the soil. A signifcant accumulation of inhibitors in the soil could occur after many precipitation events or after spring snowmelt. After a year or more, the rate of nitrogen cycling in the soil may be severely depressed. The soil collected six months after the fire treatment showed a significant increase in both nitrification and net mineralization rates. Leaching of the burned forest floor apparently stimulated nitrogen mineralization. The leachate should contain substantially lower concentrations of the inhibitors along with higher concentrations of nutrients (particularly of interest are nitrogen and phosphate). This suggests fairly frequent low-intensity fires may control the levels of inhibitors in the forest floor and soil and allow for higher rates of nitrogen mineralization.

Although nitrification potential was increased in the forest floor and eventually in the mineral soil, the field nitrate levels were only slightly increased. It appears the demand for nitrate sufficiently depletes the nitrate levels so loss of nitrate in streams should be minimal.

Conclusions

1. Low-intensity prescribed fire in ponderosa pine resulted in a slight but significant decrease in respiration in the residual forest floor material. Soil respiration was unaffected by the fire treatments.

2. Nitrogen mineralization and nitrification in the forest floor was immediately increased and remained higher 6 months after the fire treatment. The soil was not immediately affected, but both nitrogen mineralization and nitrification were significantly increased 6 months after the treatment.

3. The factor which primarily controls nitrogen mineralization and nitrification in this ponderosa pine ecosystem is allelochemic inhibition. However, it is unlikely that the inhibition was caused by tannins and polyphenols which are most commonly suggested in the literature. The inhibition is more likely caused by low temperature volatiles, such as terpenoids.

The low mineralization rates in ponderosa pine ecosystems are probably the result of fire restriction. In the absence of fire, the low temperature volatiles may build up to inhibitory levels. Periodic low-intensity fires would probably consume or volatilize the inhibitors and result in greatly increased rates of nitrogen mineralization. The mineralization of other nutrients (particularly phosphate) may also be increased.

Table 1.1. Nitrogen mineralization potentials for ponderosa pine soil (0-10 cm depth) and amended soil. Values are net amount present in soil after 10-week incubation at 20°C. Values in parentheses are amounts added in the amendment. All values are expressed as μg N g^{-1} dry weight soil (Gosz and White, unpublished data).

Amendment	NH_4-N	NO_3-N	Σ	Net (Σ minus added)
Water	8.8 b* (0)	1.2 c (0)	10.0 c (0)	10.0 a
Water + NH_4-N	12.8 ab (5.5)	3.5 b (0)	16.3 ab (5.5)	10.8 a
Water + NH_4-N + Inoculum	10.3 ab (6.9)	10.5 a (2.1)	20.8 a (9.0)	11.8 a
Water + NH_4-N + Sterilized Inoculum	14.7 a (6.8)	4.7 b (2.1)	19.4 ab (8.9)	10.5 a
Water + Nutrient Solution (complete minus N)	13.0 ab (0)	1.9 c (0)	14.9 bc (0)	14.9 a

*Values within a column with different letters are significantly different ($P<0.05$).

Table 1.2. Comparison of heterotrophic respiration measurements for the control (C), pre-treatment (B), and post-treatment (T) plots. All data are means of weekly CO_2 levels (mg CO_2 g^{-1}). For each date, means in the same column followed by different letters are significantly different ($P<0.05$). The prescribed fire treatment was performed on November 7, 1983.

		Respiration	
Forest floor	Plots	Dry Weight Basis	Ash-free Weight Basis
May 19, 1982	B	8.40 a	11.82 a
	C	8.50 a	11.86 a
August 5, 1982	B	7.55 a	15.10 a
	C	7.41 a	13.10 a
November 7-8, 1983	B	7.98 a	13.47 a
	C	7.43 a	12.70 ab
	T	4.18 b	10.77 b
May 15, 1984	C	8.61 a	14.6 a
	T	5.18 b	11.2 b
August 31, 1984	C	9.48 a	18.4 a
	T	5.28 b	11.0 b
Soil (0-10 cm depth)			
May 19, 1982	B	0.287 a	4.48 a
	C	0.344 b	6.00 b
August 5, 1982	B	0.237 a	5.17 a
	C	0.246 a	5.99 a
November 7-8, 1983	B	0.276 a	4.77 a
	C	0.250 a	4.56 a
	T	0.285 a	5.53 a
May 15, 1984	C	0.277 a	5.75 a
	T	0.626 b	5.59 a
August 31, 1984	C	0.222 a	4.54 a
	T	0.216 a	4.26 a

Table 1.3. Nitrogen mineralization potentials for forest floor from the control (C), pre-treatment (B), and post-treatment (T) plots. All data are means of weekly values (μg N g^{-1}). For each date, means in the same column followed by different letters are significantly different ($P<0.05$). The prescribed fire treatment was performed on November 7, 1983.

		Dry Weight Basis			Ash-free Weight Basis		
Collection Date	Plots	NH_4-N	NO_3-N	Σ	NH_4-N	NO_3-N	Σ
May 19, 1982	B	30.9 a	6.12 a	37.0 a	42.2 a	8.06 a	50.3 a
	C	32.9 a	0.80 a	33.7 a	45.8 a	1.12 a	46.9 a
August 5, 1982	B	22.9 a	2.98 a	25.9 a	46.4 a	6.04 a	52.4 a
	C	21.3 a	3.09 a	24.4 a	36.4 b	6.24 a	42.6 a
November 7-8, 1983	B	26.8 b	0.62 a	27.5 b	46.4 b	1.10 b	47.5 b
	C	38.9 b	2.99 b	41.9 b	77.0 b	6.20 b	83.2 b
	T	74.3 a	21.14 a	94.5 a	202.7 a	59.8 a	262.5 a
May 15, 1984	C	26.7 a	1.54 b	28.2 b	42.8 b	2.27 b	45.0 b
	T	41.4 a	41.5 a	82.9 a	93.7 a	94.99 a	188.7 a
August 31, 1984	C	26.7 a	2.86 b	29.6 a	53.2 a	5.99 b	59.3 a
	T	32.9 a	20.4 a	53.3 a	68.5 a	45.1 a	113.5 a

Table 1.4. Nitrogen mineralization potentials for 0-10 cm depth mineral soil from the control (C), pre-treatment (B), and post-treatment (T) plots. All data are means of weekly values ($\mu g\ N\ g^{-1}$). For each date, means in the same column followed by different letters are significantly different ($P<0.05$). The prescribed fire treatment was performed on November 7, 1983.

		Dry Weight Basis			Ash-free Weight Basis		
Collection Date	Plots	NH_4-N	NO_3-N	Σ	NH_4-N	NO_3-N	Σ
May 19, 1982	B	4.50 a	8.57 b	13.07 b	66.2 a	132.8 b	199.0 b
	C	4.42 a	11.50 a	15.92 a	80.1 a	200.5 a	280.6 a
August 5, 1982	B	2.81 a	7.36 a	10.18 a	62.2 a	167.8 a	230.0 a
	C	3.20 a	8.67 a	11.87 a	66.2 a	174.4 a	240.6 a
November 7-8, 1983	B	4.70 a	5.49 ab	10.18 a	81.5 a	96.8 ab	178.3 ab
	C	4.91 a	3.73 b	8.64 a	88.9 a	65.8 b	154.8 b
	T	5.16 a	6.06 a	11.22 a	98.5 a	115.8 a	214.3 a
May 15, 1984	C	3.67 a	6.14 b	9.81 b	76.1 a	125.6 b	201.8 b
	T	4.92 a	11.82 a	16.19 a	89.2 a	205.4 a	294.6 a
August 31, 1984	C	2.91 a	9.81 a	12.72 a	59.2 a	197.0 a	256.2 a
	T	5.03 a	16.77 a	21.80 a	99.0 a	329.3 a	428.3 a

Table 1.5. Site characteristics, local climatic conditions, and calculated fire behavior characteristics for the prescribed fire treatments. Treatments were applied on November 7, 1984. Fireline intensity data were provided by the USDA Forest Service (L. Buchanan).

	Plot Number			
	2	3	6	7
Site Characteristics:				
Maximum Slope (%)	5	5	20	15
Aspect	Ridge Top	Ridge Top	East	West
Moisture (%):				
Forest Floor (F + H)	14.3	20.5	17.0	10.4
0-10 cm Soil (pre-burn)	6.6	6.3	7.0	7.0
0-10 cm Soil (post-burn)	6.4	6.3	6.9	4.8
Forest Floor Biomass (tons/acre)	14.8	8.0	7.4	16.0
Climatic Conditions:				
Time (M.S.T.)	11:10	11:30	11:45	12:00
Dry Bulb Temperature (^{o}F)	62	67	65	71
Relative Humidity (%)	43	43	42	39
Mid-flame Windspeed (mph)	4	4	4.5	5.5
Calculated Data:				
Forest Floor Consumption (tons/acre)	7.1	0.7	3.3	9.2
Fireline Intensity (BTU/ft s)	38	38	49	59
Flame Length (ft)	2.0	2.4	2.7	2.9

Table 1.6. Extractable inorganic nitrogen levels in the forest floor from control (C), pre-burn (B), and post-burn (T) plots in a ponderosa pine ecosystem. Values are g N m^{-2}.

Plots	NH_4-N	NO_3-N	Σ
T	3.41 #	0.01	3.42 #
C	1.17	0.01	1.18
B	0.74	0.00	0.74

($P<0.01$)

Table 1.7. Nitrogen mineralization potential of pre- and post-burn forest floor samples from a ponderosa pine ecosystem. All values are expressed on an ash-free weight basis (AFW). Net mineralization is the difference between the initial inorganic nitrogen levels and those levels after the 10-week incubation (initial - 10 week).

	Σ NH_4-N + NO_3-N		Net Mineralized	
	Initial	10-week	$\mu g\ N\ g^{-1}$	$g\ N\ m^{-2}$
Pre-burn	32.7	66.4	33.7	0.76
Post-burn	273.1 #	440.2 #	167.1 #	2.09 #

(P 0.01)

Table 1.8. Comparison of mean heterotrophic respiration during a 10-week incubation of amended mineral soil samples from a ponderosa pine ecosystem managed by prescribed fire. The amendments included the addition of: demineralized water (Water Blk), water extracts of burned (Burned-1) and untreated (Control-1) forest floor samples, and the extracts diluted by a factor of 5 (Burned-5, Control-5) and by 25 (Burned-25, Control-25). Means followed by different letters are significantly different ($P<0.05$)

Duncan's Multiple Range Test

(mg CO_2 g^{-1} wk^{-1})

Treatment	Mean	Grouping
Water Blk	0.664	A
Control-1	0.570	B
Burned-1	0.563	B C
Control-5	0.524	C D
Control-25	0.513	D
Burned-5	0.506	D
Burned-25	0.502	D

Table 1.9. Mean inorganic nitrogen levels ($\mu g\ N\ g^{-1}$) during the incubation for nitrogen mineralization potentials on amended mineral soil smaples from a ponderosa pine ecosystem managed by prexcribed fire. The amendments included the addition of: demineralized water (Water Blk), water extracts of burned (Burned-1) and untreated (Control-1) forest floor samples, and the water extracts diluted by a factor of 5 (Burned-5, Control-5) and by 25 (Burned-25, Control-25). For each analysis, means followed by different letters are significantly different ($P<0.05$).

Duncan's Multiple Range Test

Ammonium			Nitrate			Ammonium + Nitrate		
Treatment	Mean	Grouping	Treatment	Mean	Grouping	Treatment	Mean	Grouping
Control-1	9.10	A	Burned-1	12.2	A	Burned-1	20.9	A
Control-25	8.86	A	Burned-5	12.2	A	Burned-5	20.6	A B
Burned-25	8.81	A B	Water Blk	11.8	A	Burned-25	20.2	B C
Burned-1	8.75	A B	Burned-25	11.3	B	Water Blk	19.7	C D
Burned-5	8.42	B C	Control-5	10.7	C	Control-25	18.5	D E
Control-5	8.28	C	Control-25	10.6	C	Control-5	19.0	E F
Water Blk	7.88	D	Control-1	9.8	D	Control-1	18.9	F

Table 1.10. Characteristics of the forest floor and the resultant water-extracts collected from unburned areas (control) and areas managed by prescribed fire (burned).

	Control	Burned
Forest Floor		
Total fresh weight (g)	1818	669
Tanning capacity (expressed as tannic acid equivalents, % fresh weight)	0.14	<0.08
Water-extracts		
Tanning capacity (expressed as tannic acid equivalents, mg/ml)	<0.4	<0.4
Relative absorbance (200 to 700nm)	8210	5760
pH	4.31	7.67
Ca (µg/ml)	25.0	81.0
Mg "	5.0	12.5
Na "	2.00	5.05
K "	19.75	24.25
PO_4-P "	1.20	2.20
NH_4-N "	3.90	5.60
NO_3-N "	0.16	0.48
Volume added to 10 g soil (ml), Concentrated extracts	1.5	1.5

Table 1.11. Net nitrogen mineralization during field incubation of mineral soil samples from a ponderosa pine ecosystem. Net amounts were determined by subtraction of the starting inorganic nitrogen levels (ammonium + nitrate) from the levels at the end of the incubation period. For each field incubation period, all the plots were averaged (Mean) and expressed on the basis of a 365 day incubation period (Annually).

Field Incubation Period	Total Incubation Time (days)	Plot 1	2	3	4	5	6	7	8	Mean	Annually
19 May '82 to 15 Aug '82	77	5.32	5.48	5.37	8.28	-0.59	12.61	3.98	2.80	5.41	25.6
15 Aug '82 to 27 Apr '83	263		10.92	10.25			13.81	11.84		11.70	16.2
15 Aug '82 to 16 Sep '83	409	38.72			1.46	16.30			43.75	25.05	22.3
7 Nov '83 to 15 May '84	189	0.0	-0.1*	-0.2*	-0.4	-0.3	0.3*	-1.9*	0.4	-0.38	-0.72
15 May '84 to 31 Aug '84	108	5.03	5.14*	5.37*	8.36	4.54	18.21*	5.14*	2.63	6.80	23.0

* Post-treatment incubations on prescribed fire treated plots.

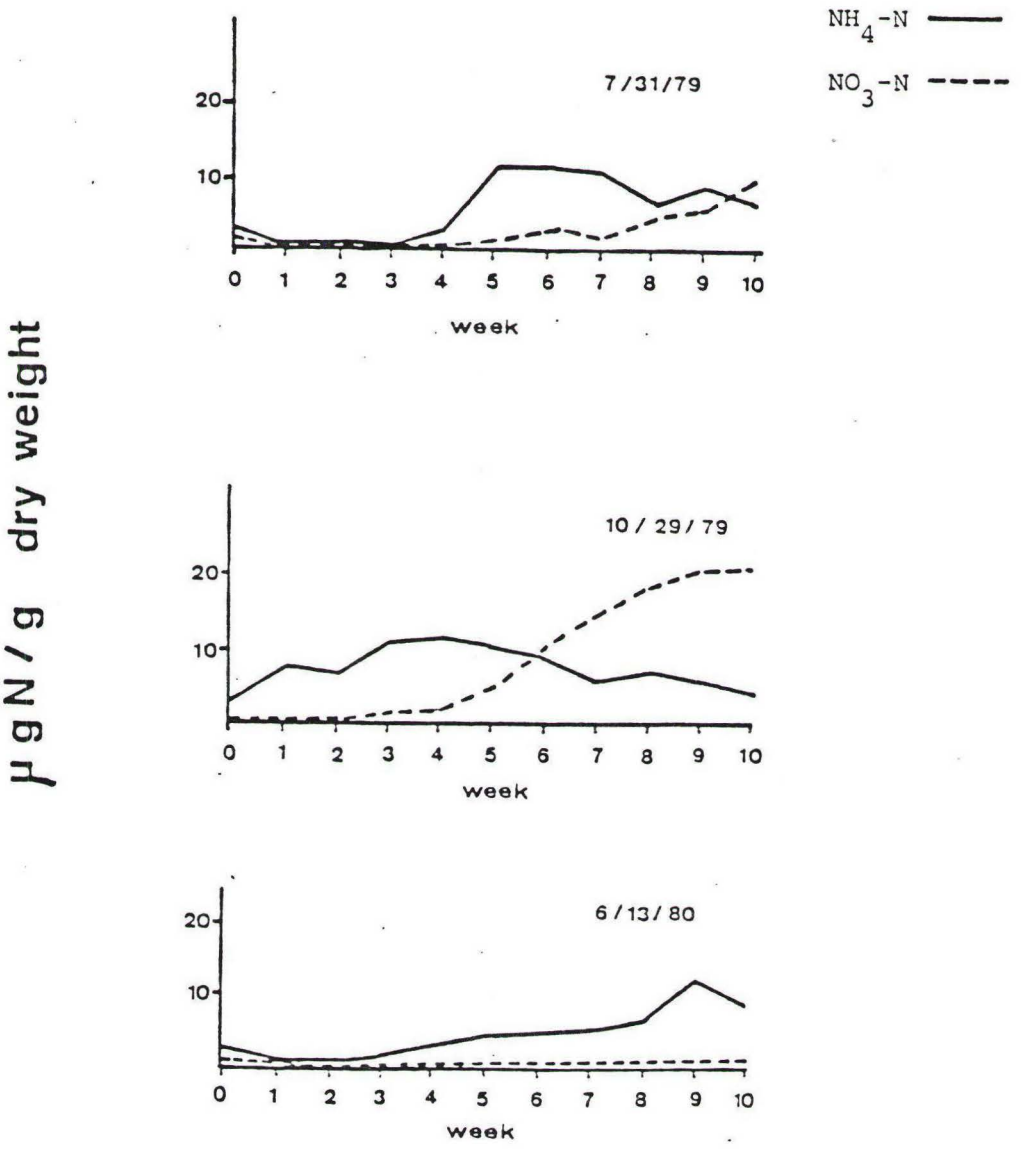

Figure 1.1. Nitrogen mineralization potential measurements on soils from ponderosa pine (described in Vitousek et al. 1982). The longest lag period is seen in the 6/13/80 collection, the shortest in the 10/20/79 collection, and intermediate in the 7/31/79 collection. Potential measurements performed on aerobic soil incubations at 20°C.

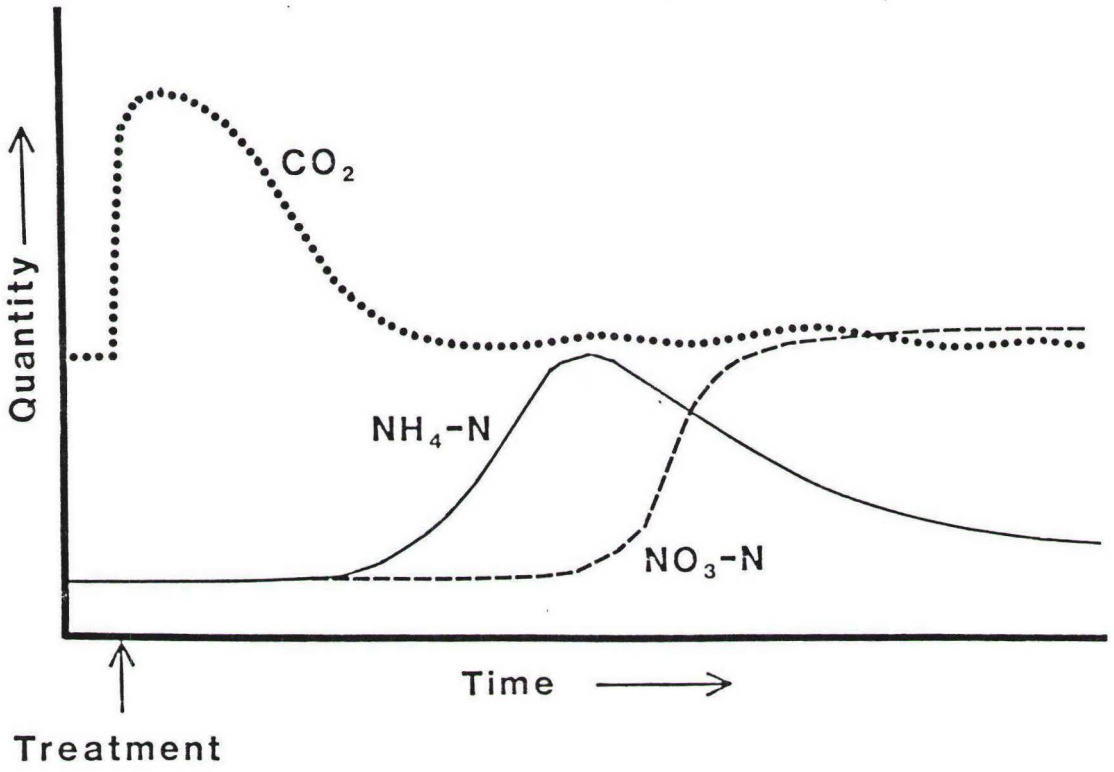

Figure 1.2. Theoretical variation in heterotrophic respiration (CO_2) and inorganic nitrogen production following disturbance (treatment). This pattern would be expected after a treatment which disturbs the forest floor or soil (such as collection of samples) and results in the creation of newly exposed or available resources to the heterotrophic decomposer community. Requirements of the decomposer community for nitrogen must be met prior to an increase in available forms of nitrogen.

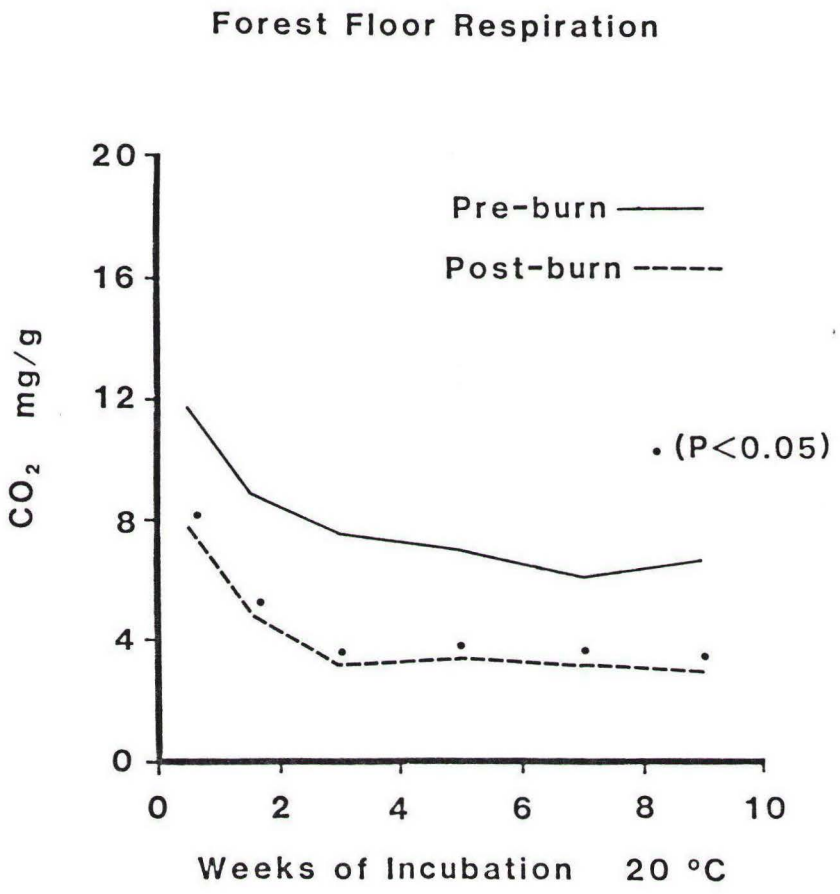

Figure 1.3. Heterotrophic respiration of immediate pre- and post-burn forest floor samples from a ponderosa pine ecosystem. Respiration is expressed as mg CO_2 evolved per gram of forest floor (including minerals and ash). Samples were collected on November 7 and 8, 1983.

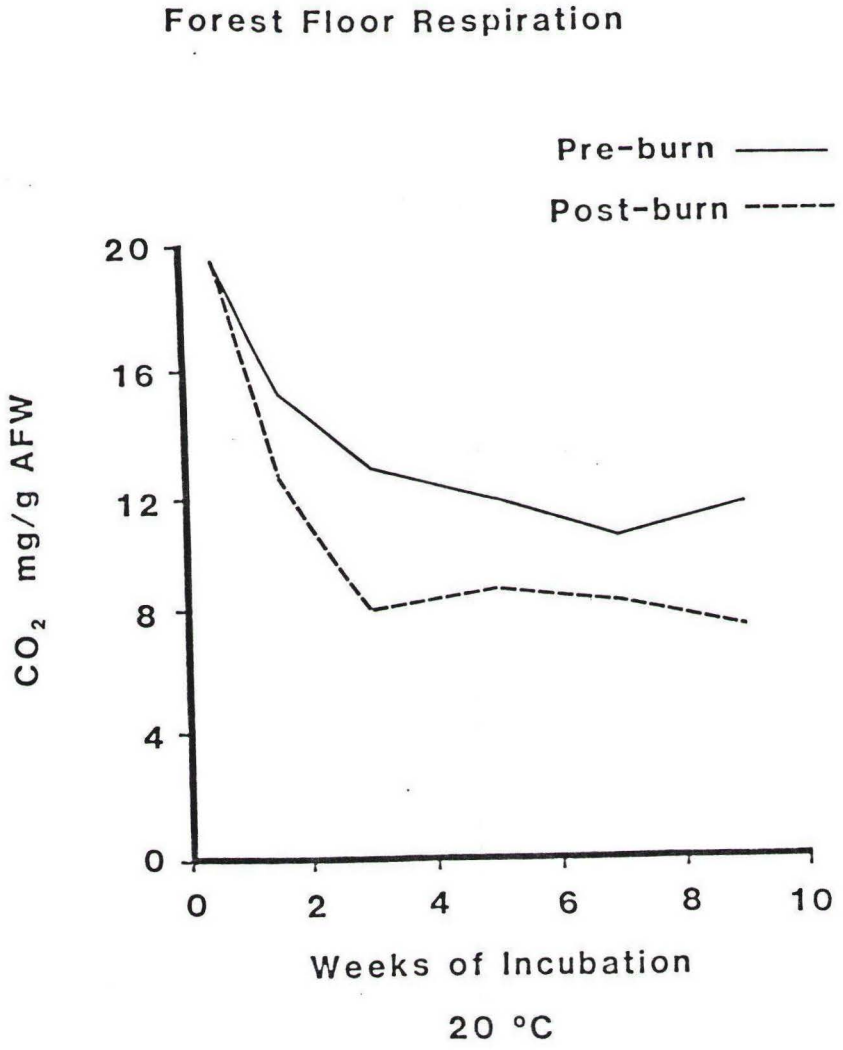

Figure 1.4. Heterotrophic respiration of immediate pre- and post-burn forest floor samples from a ponderosa pine ecosystem. Respiration is expressed as mg CO_2 evolved per gram of organic matter in the forest floor (or ash-free weight, AFW). Samples were collected on November 7 and 8, 1983.

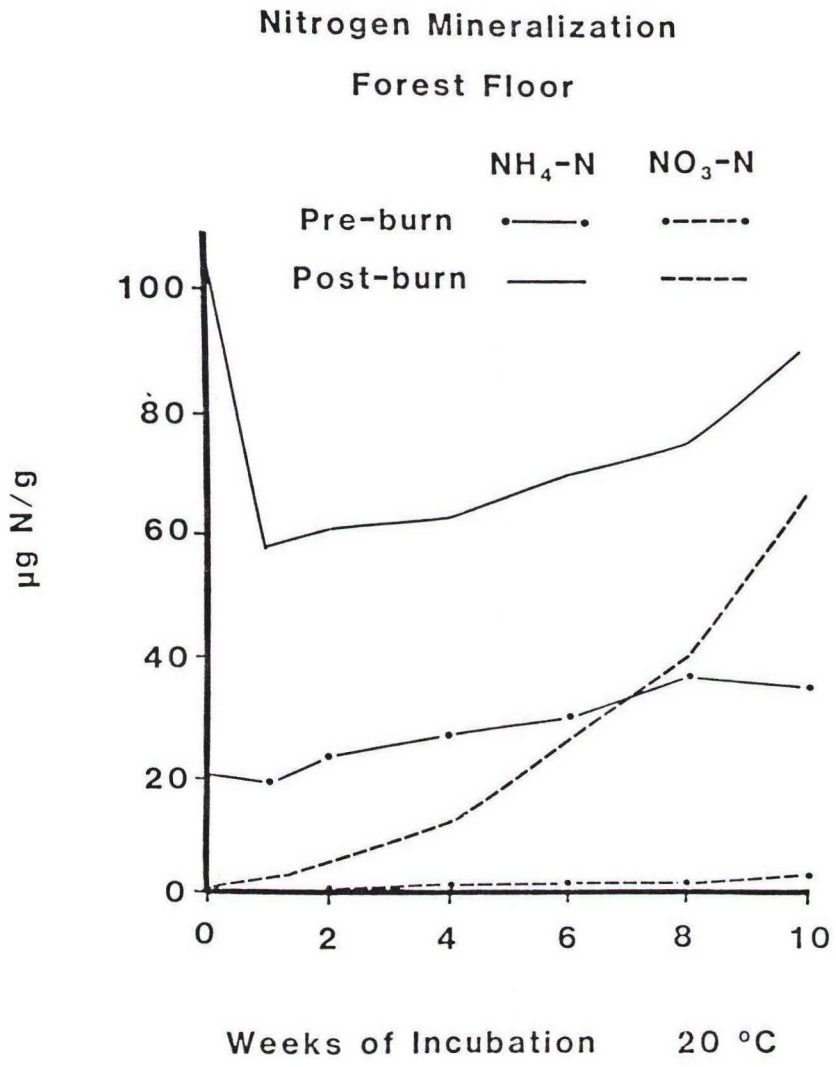

Figure 1.5. Nitrogen mineralization potentials of immediate pre- and post-burn forest floor samples from a ponderosa pine ecosystem. Values are microgram N per gram of forest floor (including minerals and ash). Samples were collected on November 7 and 8, 1983.

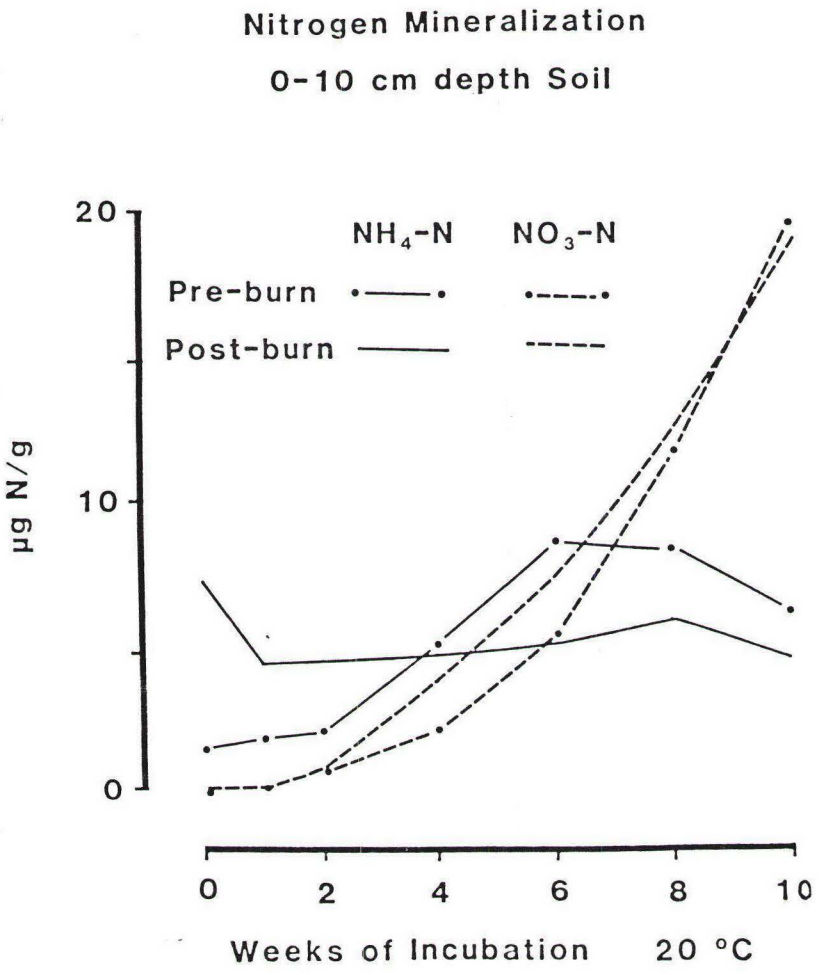

Figure 1.6. Nitrogen mineralization potentials of immediate pre- and post-burn 0-10 cm depth mineral soil samples from a ponderosa pine ecosystem. Values are microgram N per gram of mineral soil. Samples were collected on November 7 and 8, 1983.

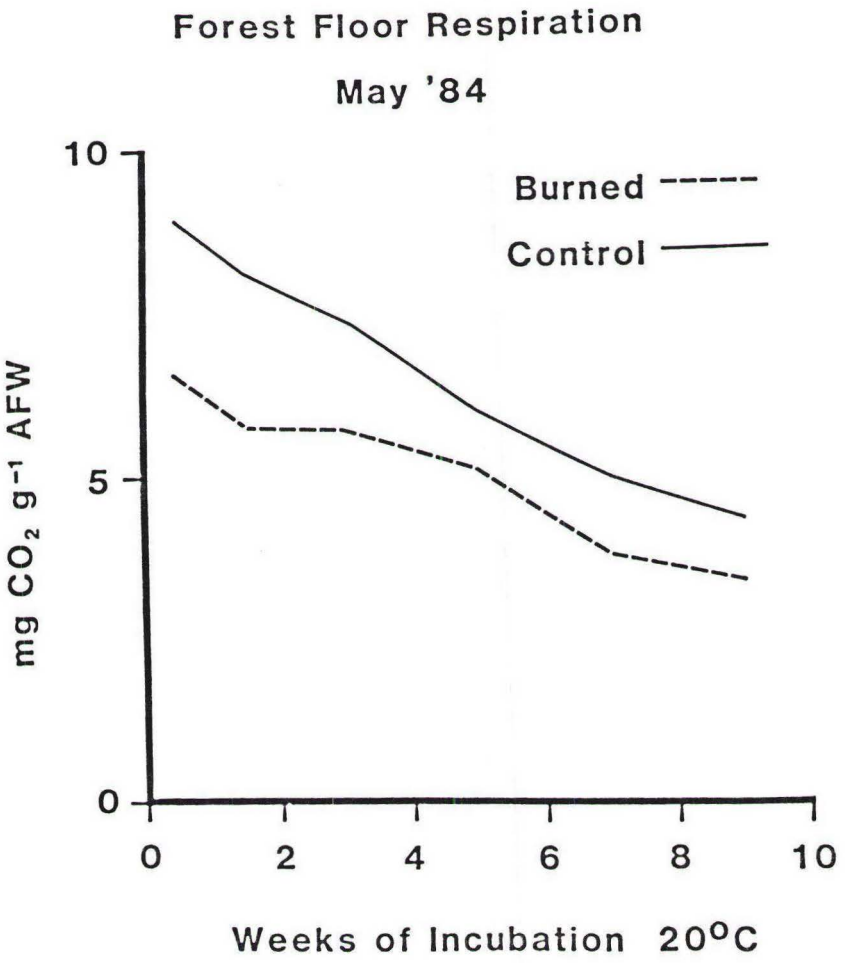

Figure 1.7. Heterotrophic respiration of forest floor samples from burned and control plots in a ponderosa pine ecosystem. Respiration is expressed as mg CO_2 evolved per gram of organic matter in the forest floor (or ash-free weight, AFW). Samples were collected on May 15, 1984.

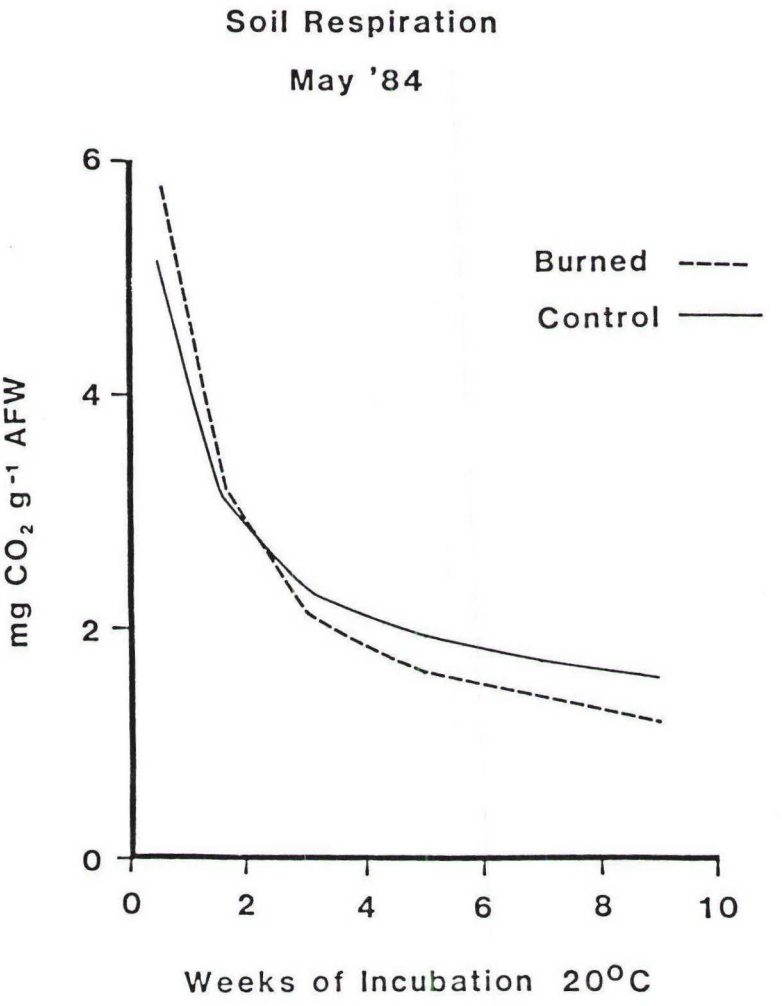

Figure 1.8. Heterotrophic respiration of 0-10 cm depth mineral soil samples from burned and control plots in a ponderosa pine ecosystem. Respiration is expressed as mg CO_2 evolved per gram of mineral soil. Samples were collected on May 15, 1984.

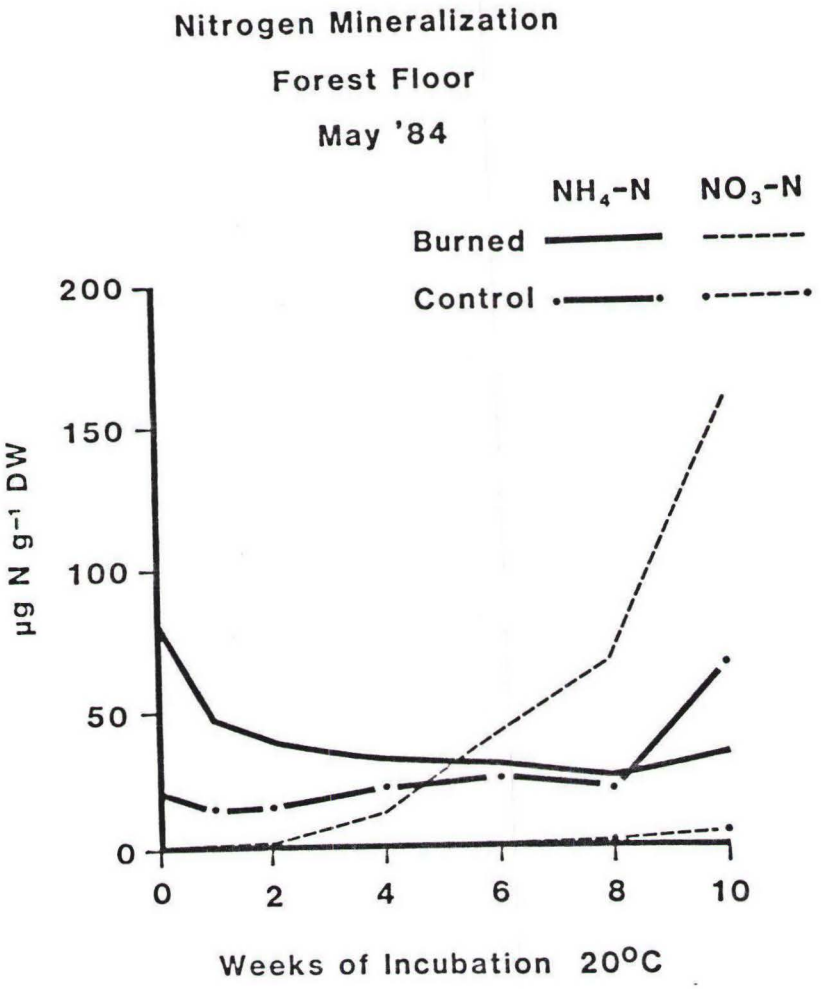

Figure 1.9. Nitrogen mineralization potentials of forest floor samples from burned and control plots in a ponderosa pine ecosystem. Values are microgram N per gram of forest floor (including minerals and ash). Samples were collected on May 15, 1984.

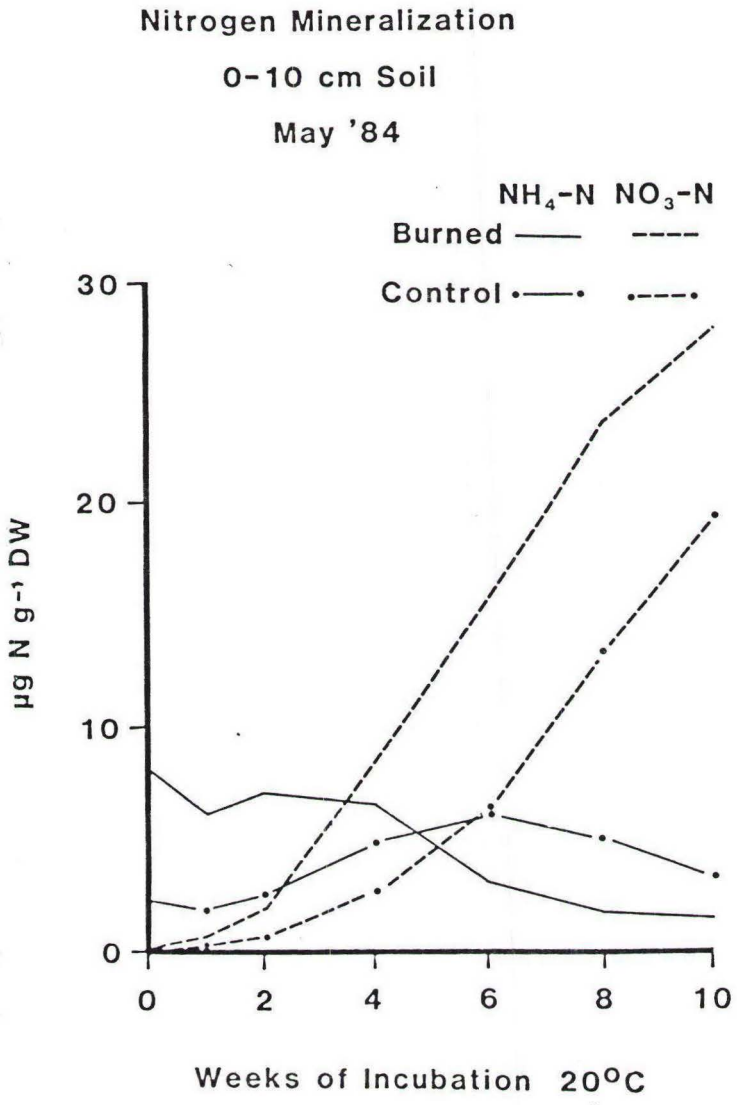

Figure 1.10. Nitrogen mineralization potentials of 0-10 cm depth mineral soil samples from burned and control plots in a ponderosa pine ecosystem. Values are microgram N per gram of mineral soil. Samples were collected on May 15, 1984.

Section 2

Effects of Fire on Organics

Introduction

Ponderosa pine watersheds in the southwest provide valuable resources including surface water for downstream users. The quality of this water is of prime importance, however, little is known about the organic quality of surface water draining ponderosa pine ecosystems. Even less is known about the effects of management with prescribed fire on organics in these waters. An objective of this section was to evaluate the effects of prescribed fire in a ponderosa pine ecosystem on water-soluble organics. An area with perennial water was not available, thus, this study considered only the water-soluble (or extractable) organics in the forest floor. The organics leached from the forest floor should determine to a large degree the organic quality of the surface waters.

It has been suggested that the forest floor of ponderosa pine contains a wide variety of inhibitors (Lodhi and Killingbeck 1980, del Moral and Cates 1971, Rice 1984). Field studies have not often supported the hypothesized allelopathic nature or the forest floor. The results in Section 1 of this report suggests the allelopathic inhibition of nitrification was removed in the forest floor by prescribed fire in this ecosystem. This section also includes the initial analyses of the forest floor samples from plot #7 collected immediately before and after the prescribed fire treatment and 5 months later. This site displayed the largest increase in nitrate production and the forest floor was used in the bioassay experiment in Section 1.

Methods

Relative organic content was analyzed by high pressure liquid chromatography (HPLC). Four g of forest floor or 10 g of mineral soil was blended with 30 ml of HPLC-grade water with an Omni mixer, suction-filtered, and a portion filtered through a 0.45 micron filter. The final filtered sample was injected into the dual solvent HPLC with water and acetonitrile as the solvents. A 24-minute isocratic period was followed by a 0 to 70% acetonitrile gradient over a 30-minute period, and finally by a 70 to 100% acetonitrile gradient over a 10-minute period.

Forest floor samples were analyzed for terpenoids and tannins by Dr. Rex Cates' laboratory. A gas chromatograph (GC) was used for the terpenoid analyses. A hemoglobin method was used to determine the tanning capacity of the forest floor.

Results and Discussion

The three samples of forest floor from plot #7 were analyzed for terpenoids by the GC method (Fig. 2.1). The nitrogen mineralization potential of the forest floor samples showed little or no nitrification in the pre-burn sample while both the post-burn samples produced large quantities of nitrate. Thus, if this analysis detects the compound(s) responsible for inhibition, the inhibitor must be absent or in reduced amounts in the two post-burn samples and present or in greater amounts in the pre-burn sample. The immediate post-burn sample had an immediate reduction in nearly all the component peaks. However, the 5-month post-burn sample showed an actual increase in some components and had other components which were not present in the pre-burn sample.

A known quantity of an internal standard (shown at retention time 17.6) was added to each sample. The area of the standard peak provided a means to convert each of the component peaks to an equivalent weight. Comparison of these equivalent amounts showed tnat all the compounds with retention times greater than 24.7 minutes had equal or greater concentrations in the post-burn samples than in the pre-burn sample. This indicates that these components were not responsible for inhibition of nitrification in the forest floor. The components with shorter retention times (and lower boiling points) were the only components with lower concentrations in both post-burn samples than in the pre-burn samples. Table 2.1 lists the retention times, relative concentrations, and tentative identification of the components with retention times less than 24.7 minutes. Only the component tentatively identified as alpha-pinene has concentrations equal to or greater than the corresponding component in the pre-burn sample. Thus, any of the other components could be responsible for the inhibition of nitrification in the pre-burn sample. The

most likely components (based on absolute concentration) are those with retention times of 8.5, 9.06, 10.7, and 12.8 minutes.

These 3 forest floor samples were also analyzed for total tanning capacity. None of the samples, including the pre-burn sample, had a tanning capacity of more than 0.1% relative astringency. This level is considerably below the levels reported for inhibition if nitrification in other ecosystems (Rice 1984). This strongly argues that tannins or tanning capacity is not the cause of nitrification inhibition in this ecosystem.

Unfortunately, the reverse phase HPLC suffered major damage during this study and the post-treatment samples have not been analyzed for water-soluble components. The pre-burn samples all had similar chromatographs. The main variation between sites was in the height of the peaks. The chromatographs appear to be rather characteristic of ponderosa pine material in this general area. Samples collected earlier at other locations (the Tesuque Watersheds and in a nearby stand in the Jemez Mountains) had very similar chromatographs (see the detailed study plan and the original proposal for other chromatographs).

The forest floor from plots #7 and #8 were extracted with water for the bioassay experiment in Section 1. The relative absorbance of the extracts (Table 1.10) showed plot #7 (burned) was only 30% lower than plot #8 (control) although the total amount of forest floor in plot 8 was nearly three times more in plot #8 than plot #7. This is probably due to the humus layer containing the majority of water-extractable organics. The humus layer was the least affected by the fire treatment. These results indicate the prescribed fire treatments may reduce the level of organics in surface water, but the reduction is much less than the reduction in forest floor biomass following prescribed fire.

Conclusions

Prescribed fire in pondedrosa pine ecosystems reduced the water-soluble organic fraction in the forest floor. This may result in lower levels of organics in streams. However, the reduction in forest floor was much greater than the reduction in dissolved organics.

The prescribed fire resulted in an immediate reduction in most all volatile organics. The greatest decrease in samples 5 months after the prescribed fire was in the lower boiling point volatiles (mainly monoterpenes and alcohols). Four compounds (identified by their retention times) have been identified as the most likely inhibitors of nitrification. The role of these volatiles and others in the control of ecosystem processes merit further investigation.

Figure 2.1. Chromatographs for the analysis by gas chromatography of the pre-burn, post- burn, and 5 month post-burn samples of forest floor from plot 7. The internal standard (fenchyl acetate) accounts for the peak at about 17.66 minutes retention time. Many of the peaks are due to the ether extract (particularly at 27.25).

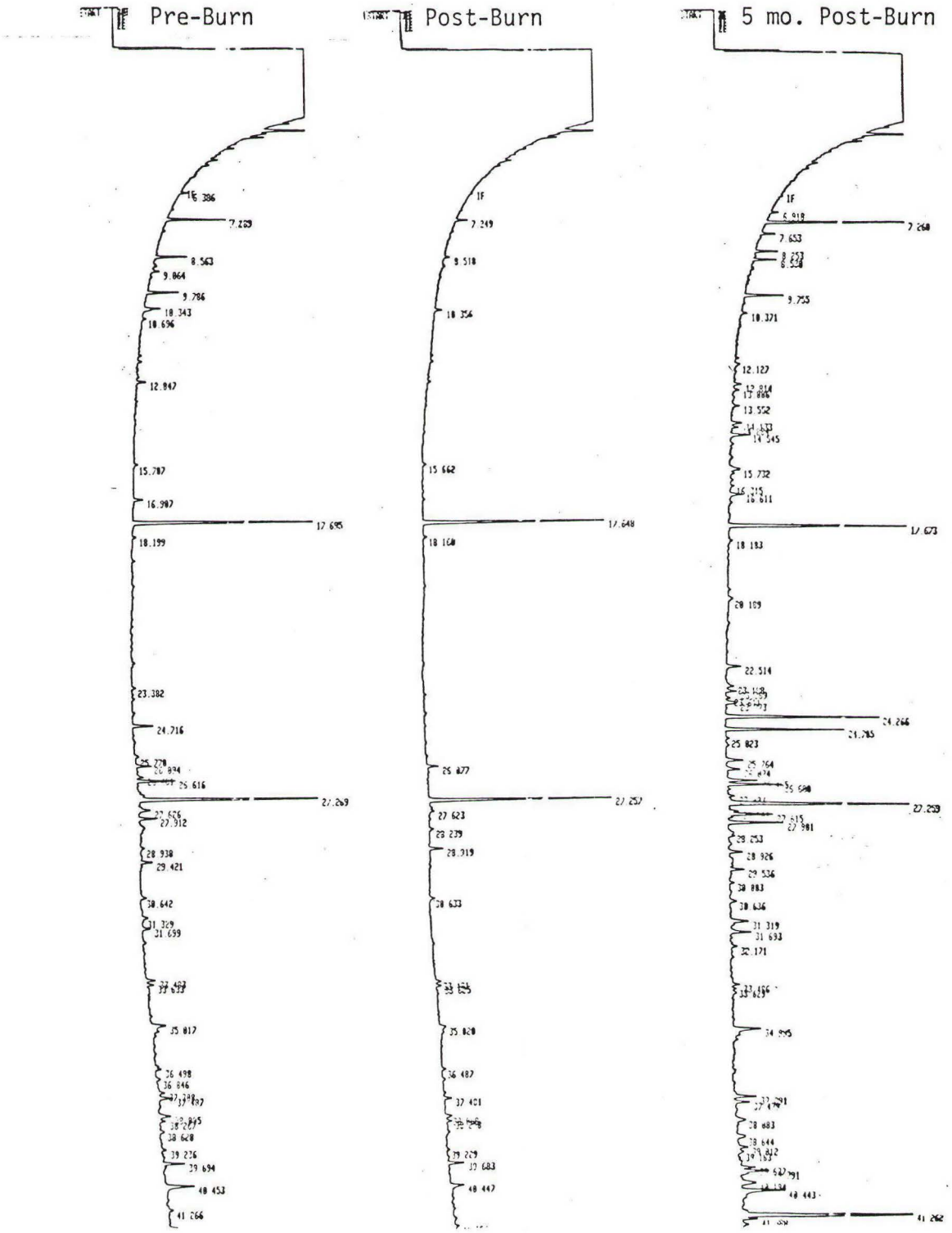

Table 2.1. Retention times (RT), tentative identification (I.D.), relative concentration (expressed as part per million of dry tissue weight) (PPM), and % of the pre-burn forest floor sample for peaks on the gas chromatographs.

I.D.	RT	Pre-Burn PPM	Immediate Post-Burn PPM	Immediate Post-Burn %	5 month Post-Burn PPM	5 month Post-Burn %
	6.4	0.0055		0		0
	6.9				0.0032	-
α-pinene	7.26	0.0454	0.0035	7.7	0.0818	180.2
camphene	7.65				0.0061	-
	8.25				0.0108	-
β-pinene	8.53	0.0251	0.0017	6.8	0.0105	41.8
	9.06	0.0056		0		0
	9.76	0.0270		0	0.0205	75.9
limonene	10.36	0.0135	0.0027	20.0	0.0055	40.7
	10.70	0.0036		0		0
	12.13				0.0029	-
	12.83	0.0098		0	0.0045	45.9
	13.01				0.0035	-
	13.55				0.0061	-
	14.13				0.0073	-
	14.28				0.0034	-
	14.54				0.0130	-
	16.32				0.0023	-
	16.70	0.0116		0	0.0087	75.0
	20.20				0.0050	-
	22.51				0.0097	-
	23.19				0.0046	-
	23.37	0.0099		0	0.0057	57.6
	23.61				0.0025	-
	23.77				0.0109	
	24.27				0.0966	-

Section 3.

Effects of Volatiles on Nitrogen Mineralization

Introduction

Decomposition of plant materials is affected by many biotic and abiotic factors. In coniferous forests, tannins and polyphenols are suspected to inhibit the rate of decomposition (Gosz, 1981). These compounds bind with easily decomposed materials and form resistant complexes. Many of these same compounds have been reported to be toxic to nitrifying bacteria (Lodhi and Killingbeck 1980). Lodhi and Killingbeck attributed the lack of nitrate in soil from a ponderosa pine ecosystem to the occurence of toxic water-soluble tannins and other polyphenols leached from the ponderosa pine needles and litter.

Our research does not support aspects of this hypothesis. First, the residual forest floor collected within 24 hours following a prescribed fire displayed increased ammonification and nitrification rates during incubation (see Section I, Effects on Nitrogen Mineralization). The increase in nitrate means the nitrifyers were present in the forest floor before the treatment, because it is unlikely they could have been introduced that quickly after the prescribed fire. If the tannins present before the fire were "toxic", nitrifying bacteria would have been present in very low numbers or not at all. Secondly, soil from the hottest prescribed fire (plot #7) had significantly increased nitrogen mineralization and nitrification rates. Direct combustion of the tannins and polyphenols in the soil is highly unlikely since the fire did not consume the entire forest floor. However, compounds with low volatilization and/or boiling points, including some polyphenols, may have been driven out of the soil or decomposed by the heat pulse in the soil.

An alternative hypothesis to explain the above results is: Volatile organics (e.g., terpenoids) produced by ponderosa pine inhibit ammonification and nitrification. The volatile materials are not directly toxic. Volatile organics have been reported to inhibit microbial species in one or more of the following manners: 1) reduced spore germination, 2) causing ceased spore formation, 3) reduced mycelia growth, and 4) promotion of or release from fungistasis (Stotzky and Schenck 1976). The source of the volatiles would be the fresh litter and the forest floor. Transport of the volatile organics could be in the aqueous phase and in the volatile phase. Many terpenoids are slightly water-soluble and could be transported in the aqueous phase. Stream water from a spruce-fir watershed was found to have 75% of the dissolved organics in the neutral fraction consisting of terpenoids (Rex G. Cates, personal communication). Volatile organics would migrate along a thermal gradient from the warmer regions to the cooler regions where condensation of the volatiles could occur. Assuming heating of the forest floor and soil occurs by solar insolation, such a thermal gradient within the forest floor and soil could be established on nearly a daily basis.

Volatiles are often consumed early in the pyrolysis process. Terpenoids have low boiling points and can form flammable volatile mixtures well in advance of the flame front in a fire (Chandler et al. 1983). If the terpenoids (and/or other volatiles) are responsible for the inhibition of nitrification and/or ammonification, this would be consistent with our results and explain how fire initiated such an immediate increase in mineralization and nitrification.

Literature Review

The effects of volatile organics on the process of decompositon are not well known with very little written on the subject. Since the process is primarily performed by microbes, the literature on the effects of volatiles on microbes would be applicable to such an investigation. Unfortunately, this information is equally sparce. Muller (1965) noted smaller bacterial populations in the soil of the bare zones beneath the shrubs of the chapparal area than in the grass zone. He suggested this may be a result of inhibition by volatiles in the bare zones, or a stimulation of growth in the grass zones by organic debris from the grasses.

The terpenoids and other volatiles have been studied mostly with respect to their role in resistance to plant pathogens. Resistance to bark beetle has been attributed to the reduced growth of the associated fungi due to volatiles and other constituents of the tree resins (Raffa and Berryman 1982; Smith 1977; Cobb et al. 1968; and Sturgeon 1979). The majority of this work deals with the effects on pure cultures of fungi after exposure to select terpenoids or resins collected by tapping the tree. The effect of a given terpenoid varied between the species of fungi. Some fungal species were inhibitied while others were stimulated in their growth. A wide range of responses were displayed to different terpenoids dependent upon the fungal species. Thus, there is no apparent agreement about the effects of a specific terpenoid on a mixed microbial community in a soil.

Inhibition of the processes of ammonification and nitrification may occur (and probably does occur) by the combination of more than one chemical substance. Also, while a given compound may inhibit one group of organisms (i.e., the decomposers), another group may be able to utilize the coumpound as

an energy source. Every known naturally occuring compound will eventually be consumed by some microorganism. Thus, the effects of any compound will be transient. This transient effect may account for the reported seasonal patterns and some of the apparent discrepancy in the literature.

Experimental Methods

There are no established methods applicable to testing the influences of volatiles on soil microbes and their processing rates. The following experiments contain aspects of several methods used by different researchers. The proportion of forest floor to mineral soil which has been measured in the field was maintained in the experiments.

I. Thermal Gradient.

Forest floor material was placed in a glass flask and submerged in an oil bath. This flask was connected by a glass tube to another flask which contained mineral soil from a burned site (the soil had higher rates of nitrification than unburned soil). The flask with the forest floor was heated to 100^{o}C. The other flask with mineral soil was kept cool in an ice-water bath. The tube connecting the two flasks was covered with an alumimum foil tent. The tent was to keep the glass warm and aid in the transfer of volatile gases from the forest floor to the mineral soil. The forest floor was heated in the oil bath for 48 hours. After this period, the heated forest floor , the receiving mineral soil, and an untreated mineral soil sample were then analyzed for heterotrophic respiration and nitrogen mineralization potential.

II. Temperature Gradient.

From an homogenous forest floor sample, subsets of 120 grams were heated at room temperature (about 25), 40, 60, 80, and 100^{o}C for 48 hours. After heating, the forest floor samples were adjusted for moisture content and inoculated with soil capable of nitrification (5% inoculum by weight). The forest floor subsets were proportioned into three jars. Each jar was analyzed for respiration and mineralization potential.

III. Trapped Vapor.

Six (6) replicate jars of mineral soil from a burned plot (the same mineral soil as in experiment II) were used for each of the following amendments: no treatment (control), added forest floor, and added terpenoids to simulate ponderosa pine resin. The forest floor material or terpenoids were placed in small beakers and set inside the jar to avoid contact with the soil except by vapor. Three (3) of the replicates were moisture corrected, the jars sealed, and placed in the dark at room temperature. The other 3 replicate jars were sealed, set in an oven at 60°C for 6 hours (to simulate heating of the material under field moisture conditions), removed and cooled. The soil moisture was adjusted and the jars were treated in the same manner as the first 3 replicates. All jars were opened weekly to replenish their atmosphere and maintain aerobic conditions. After 10 weeks, the soil was analyzed for nitrate and ammonium by extraction with KCl (see methods in Section 1).

The terpenoid treatment consisted of equal parts of limonene, myrcene, alpha-pinene, beta-pinene, and beta-phellandrene. This mixture contained the major terpenoids found in ponderosa pine resin (Cobb et al. 1968). This terpenoid addition may identify the influence of these terpenoids versus other volatiles contained in the forest floor on the decomposer community.

Results

I. Thermal Gradient:

The apparatus used in the thermal gradient experiment to transfer volatile gases from the forest floor to the mineral soil was not effective. Volatiles condensed in the glass connecting tube. Thus, many volatiles never reached the soil or were greatly reduced in concentration. Not too suprisingly, the respiration and nitrogen mineralization rates in the experimental soil and the control soil were not significantly different. The apparatus must be modified to provide an adequate test of this mechanism of transfer. The flask containing the forest floor could be modified to allow entrance of a stream of air. This airflow may assist in moving the volatiles through the connecting tube and to the soil. Another modification may include placing the forest floor on a screen above the soil and heating the forest floor from above by a heat lamp.

II. Temperature Gradient:

The initial heating of the forest floor increased the initial ammonium concentrations in the forest floor in direct proportion to the temperature. This increase is generally attributed to the mortality of soil organisms. Within the first 2 weeks of incubation, all the forest floor samples immobilized ammonium resulting in nearly equal concentrations of ammonium in all samples. After the immobilization phase, no mineralization of ammonium or nitrate occurred in any of the forest floor samples. This may indicate microbial demand for nitrogen was not satisfied during the incubation period.

The effects of the temperature gradient on respiration are shown in Table 3.1. The forest floor heated in the oil bath was included in this analysis. The intermediate temperatures significantly increased respiration,

according to these data. However, these data are potentially misleading. Visual inspection of the forest floor material suggested an increase in microbial biomass (both hyphae density and bacterial colony size) with increasing temperature (the only exception was the oil-bath treated forest floor). Also, changes in the composition of the microbial community may have occurred. A similar increase in microbial biomass was noticed in earlier experimental trials. If large increases in microbial biomass occurred, respiration data alone would not accurately indicate the amount of substrate consumed by the decomposer community. Changes in the size and composition of the heterotrophic community would alter amount of carbon respired in proportion to the amount of substrate consumed.

Both the observed size of the colonies and the immobilization of nitrogen in the nitrogen mineralization potentials strongly suggest an increase in available substrate occurred after heating. Increase in substrate availability would account for the immediate immobilization of nitrogen in actively growing microbial populations. The continued growth of the microbial populations during the incubation period would result in continued demand for nitrogen and could account for the lack of mineralization during the incubation period.

To further examine the above observations, this experiment should be performed utilizing smaller amounts of forest floor per container. Also, microbial biomass measurements should be performed at the end of the incubation period. Microbial nitrogen can also be measured with this method. These data would be required to properly evaluate the temperature gradient experiment.

III. Trapped Vapor:

These results were particularly interesting. Heating the jars and their contents had a significant effect on nitrogen mineralization and nitrification (Table 3.2). All the jars which were heated at 60^{o}C showed a dramatic decrease in nitrate production. These soils were not reinoculated after heating. This indicates the nitrifying populations were particularly susceptible to heat effects. Nearly all of the mineralized nitrogen remained in the ammonium form in the heated soils. The soils exposed to the forest floor were not significantly different from the control soils for ammonium, nitrate, or the net amount of inorganic nitrogen. Exposure to the terpenoids resulted in significantly lower ammonium levels and net inorganic nitrogen levels. The terpenoids completely inhibited nitrification where the other heated soils had detectable amounts of nitrate.

When the effects of trapped gases on the room temperature soils were evaluated, greatly different results occurred. The control soil was particularly efficient at converting ammonium to nitrate and had the highest net inorganic nitrogen production. The forest floor vapor significantly reduced the net mineralization (the lowest of all samples). The forest floor did not effectively inhibit the process of nitrification with nitrate about equal to ammonium. The terpenoids lowered net mineralization, but not as much as the forest floor. However, the terpenoids were 100% effective at inhibiting nitrate production (100% inhibition occurred at both temperature exposures).

Discussion

The sensitivity of the nitrifying bacteria to the 60°C exposure was unexpected. Belser (1979) cited studies where incubation of soils at 40°C completely inhibited nitrification and other studies where measured nitrification occurred in soils up to 60°C. The general conclusion was indigenous nitrifiers had temperature optima and maxima adapted to their climatic region. Ponderosa pine ecosystems are generally thought to be warm, dry forests. However, it is doubtful the mineral soil in ponderosa pine would ever be exposed to these temperatures (except where no forest floor exists) under normal field conditions. Soil temperatures may reach this level and hotter during prescribed fires and wildfires. The effects of the duration of exposure to the elevated temperature (6 hours) and the water content of the soil on the mortality of the nitrifying populations are unknown. Dunn and DeBano (1977) reported that Nitrosomonas and Nitrobacter bacteria were killed in dry soil at temperatures of 140°C, but at only 75 and 50°C, respectively, in wet soil. Although the assay soils were at field moisture content, perhaps more or less water in the soil would alter these results.

The other results from the trapped vapor experiment were particularly interesting and unexpected (at least the magnitude of the results). These data suggest: 1) the forest floor contains vapors which inhibit nitrogen mineralization processes in the soil, and 2) the transfer of the vapor from the forest floor to the soil is very effective at room temperature. No real thermal gradient was established since the forest floor and soil were at room temperature within the same jar. Trapping the vapors within the jar may greatly increase the concentration of the vapor over concentrations which may occur in the field. The important aspect of these results is the vapor can be

liberated at what was thought to be low temperatures (room temperature). This may explain why the forest floor exposed to room temperatures for 48 hours in the temperature gradient experiment (experiment I) showed an unexpected growth of microbial colonies. It appears the loss of vapors resulted in increased growth rates of microbial populations. Thus, removal of the inhibitory properties of the forest floor may be achieved at relatively low temperatures.

The effectiveness of the inhibition by forest floor vapors apparently was greatly reduced, if not completely eliminated, in the heated jars. This suggests: 1) thermal decomposition of the inhibitor occurred; or 2) the inhibitor was denatured (altered configuration of the molecule); or 3) the inhibitor was irreversibly bound to another substrate; or 4) the source of the inhibitor (a specific microbial species) was destroyed during the heating period. The possibility that a volatile inhibitor was being produced by a microbial population can not be excluded. Changes in species composition of the microbial community in both the soil and forest floor within the heated jars were definitely possible. Growth of a fine filamentous fungi occurred in the soil in the heated jars with terpenoids although the filaments were not visable in the room temperature soils.

Respiration was measured on another set of jars containing the assay soils and the ponderosa pine terpenoids. Greatly increased respiration measurements in these jars indicate a portion of the microbial community can utilize the terpenoids as a carbon source. Interpretation of these data was not clear because the terpenoids partially neutralized the alkaline trap. Also, the terpenoids resulted in a decrease in soil pH by a full pH unit. These factors tend to make interpretation of the data questionable. Again, measurement of microbial biomass at the end of the experiment should determine

the amount of substrate incorporated into microbial biomass and provide insight into the effect of the terpenoids on the general decomposer community.

Conclusions

As usual with an area that has received little if any research, problems arose with the methodology. These problems are not insurmountable. The addition of microbial biomass and microbial nitrogen determinations would greatly aid in the interpretation of the effects of volatiles. The experiments should be repeated with smaller quantities of forest floor and terpenoids. This would decrease the undesirable side-effects (changes in soil pH) and increase the sensitivity of the measurements.

Even with the problems associated with the data, certain conclusions can be drawn. 1) Volatiles and terpenoids in the forest floor of ponderosa pine ecosystems can inhibit the processes of nitrogen mineralization and nitrification. 2) Volatiles can influence soil properties at relatively low temperatures (20^{o}C or lower). Whether the concentration of volatiles or terpenoids in the trapped vapor experiments would ever occur in the field is questionable, however, their role of volatiles (particularly terpenoids) in the control of these ecosystem-level processes merits further attention.

Many ecosystems which display low rates of nitrogen mineralization are those which historically were prone to frequent fires. The low mineralization rates may be the result of the restriction of fire. The restriction of fires in these ecosystems may allow the accumulation of volatiles to inhibitory levels. The use of prescribed fire in fire-prone ecosystems may increase the nitrogen cycling (and possibly the productivity) of these ecosystems.

Table 3.1. Respiration from forest floor samples incubated at 20°C after being heated for 48 hours at: room temperature (Rm Temp), 40, 60, 80, or 100°C, or heated in an oil bath at 100°C (100°C OIL). Means followed by different letters are significantly different ($P<0.05$).

Duncan's Multiple Range Test

(mg CO_2 g^{-1} wk^{-1})

Treatment	Mean	Grouping
60°C	8.51	A
80°C	8.47	A
40°C	8.44	A
100°C	7.99	B
Rm Temp	7.59	B C
100°C OIL	7.47	C

Table 3.2. Inorganic nitrogen levels ($\mu g\ N\ g^{-1}$) in assay soils after 10-week incubation at 20°C. Vials containing either forest floor samples (FF), simulated ponderosa pine terpeniods (Trep), or nothing (Con) were placed in the jars with the assay soil and heated at room terperature (Rm) or 60°C for Six hours prior to incubation. For each analysis, means followed by different letters are significantly different ($P<0.05$).

Duncan's Multiple Range Test

Treatment	Mean	Grouping
	Ammonium	
FF (60°C)	47.2	A
Con (60°C)	43.3	A
Terp (60°C)	27.7	B
Terp (Rm)	20.6	C
FF (Rm)	9.1	D
Con (Rm)	0.8	E
	Nitrate	
Con (Rm)	62.9	A
FF (Rm)	7.9	B
Con (60°C)	2.5	C
FF (60°C)	0.6	C
Terp (Rm)	0.0	C
Terp (60°C)	0.0	C
	Ammonium + Nitrate	
Con (Rm)	63.7	A
FF (60°C)	47.9	B
Con (60°C)	45.8	B
Terp (60°C)	27.7	C
Terp (Rm)	20.6	C D
FF (Rm)	17.0	D

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